METABOLISM OF SILAGE DIETS

IN THE RUMEN

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by

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SUMMARY

Four separate experiments were carried out to investigate the nutritive value of silages, by balance trials and in terms of certain rumen fermentation characteristics. The first two experiments were concerned with the establishment of satisfactory methods of characterising rumen contents. Another compared silages made by different silo-filling techniques and the last examined the effect of pre-wilting and prewilting plus the addition of formic acid.

The work offered an opportunity of comparing the fate in the rumen of dried and fresh grass, and autumn and spring grass.

In the feeding regime finally adopted, food was given in two discrete meals per day, at intervals of twelve hours, with access allowed for two hours. This gave a definite pattern of fermentation activity after ingestion of food, as measured by ruminal pH, ammonia and volatile fatty acids. The patterns obtained during daylight did not differ from those during the night and were reproducible as long as conditions remained constant. There was considerable between animal variation. Increasing the frequency of feeding, or allowing free access to the food, reduced the range of values of the rumen parameters. Additionally, free access resulted in erratic feeding behaviour and a less definite rumen fermentation pattern. Both these modified techniques reduced the likelihood of showing significant differences.

Silages made with delayed sealing of the silo (treated) were compared with silages made with immediate sealing (control). Treated silages had higher intakes of metabolisable energy and protein despite reductions in digestibility. When the silages made from first cut material were fed, treatment did not affect ruminal pH, total volatile fatty acid or ammonia concentrations or their curve patterns. An increased proportion of ruminal propionic acid, giving a narrower ratio of acetic to propionic acid, was obtained when treated silage was fed. When silages made

from third cut grass were used, ruminal total volatile fatty acid concentrations were higher and pH values lower for the treated silage. Curve patterns of these were again similar. The ratio of the molar proportions of acetic to propionic acid was much wider for the treated silage owing to the different curve pattern with the latter acid.

Comparison of dried with fresh grasses showed higher fermentation activity for the dried material, with a higher proportion of propionic acid, particularly with the high water soluble carbohydrate first cut material. The lack of agreement between the rumen fermentation characteristics for the fresh and dried material precluded the use of the latter in place of fresh grass for comparative purposes. Spring, compared with autumn, grass showed higher dry matter intakes, more active rumen fermentation and a higher proportion of propionic acid in the rumen volatile fatty acids.

Reduced in-silo fermentation, resulting from prewilting or prewilting plus the addition of formic acid, increased intake of dry matter compared with untreated silage, but only with the combined treatment was the intake of metabolisable energy and protein increased. Ruminal pH, total volatile fatty acid concentrations and the molar proportions of acetic acid were similar for the silages. Propionic acid values were higher for the acid treated wilted material. Ruminal ammonia concentrations were highest with the acid treated and lowest with the fresh silage. A more desirable pattern of fermentation activity was obtained with the grass than any of the silages.

CONTENTS

INTRODUCTION

Silage is the product formed when grass or other high moisture materials are conserved anaerobically by a process of controlled fermentation. Preservation results from the acidity developed by fermentation of carbohydrate to lactic acid. Formic acid is sometimes used as an additive to improve the likelihood of achieving a satisfactory preservation, by bringing about an initial increase in acidity and encouraging further increase, by lactic acid fermentation, to a situation where the mass preserves satisfactorily.

The composition of a silage depends upon the source material and upon the nature and extent of changes taking place during ensilage. Owing to these changes silage dry matter contains a greater or lesser proportion of the products of fermentation and as such differs from that of the more conventional foods.

In assessing energy sources for animals three things are important. The first is the energy content, second the availability of the energy to the animal and thirdly the efficiency with which the available energy is used by the animal. The energy value of silage dry matter might well be expected to be different, and there is some evidence to show that it is higher than the dry matter of conventional foods.

The ensiling process involves considerable breakdown of the parent material to simpler compounds. These might be expected to be more amenable to digestion and the silage to have a more digestible dry matter and a higher metabolisable energy.

Acetic, propionic and butyric acids, which are the main products of fermentation of the carbohydrate fraction of foods in the rumen, are used with quite widely varying efficiencies for various functions in the animal. The efficiency of utilization of the metabolisable energy of a food therefore depends upon the function to be performed and the proportion of the three major volatile fatty acids in the rumen contents of animals given a food. A considerable amount of information is available concerning the pattern of ruminal volatile fatty acids characteristic of a large number

of foods and mixtures of foods. It is therefore possible to predict the efficiencies with which their energy will be used by the animal and this helps in the assessment of their nutritive value. There is little information on this aspect of silage feeding. It is not possible to predict what the ruminal volatile fatty acid pattern might be since the nature of silage dry matter is so different from that of conventional foods, produced as it is by anaerobic fermentation. When silage is fed to animals, there is the unusual situation of the products of anaerobic fermentation being subjected to further anaerobic fermentation in the rumen, to provide an energy source for the animal.

In assessing nitrogen sources for animals it is necessary to know the nitrogen content, the availability to the animal and the efficiency of utilization of the available nitrogen by the animal.

The nitrogen content of silages is, generally speaking, the same as or frequently higher than that of the parent material. Ensilage involves extensive breakdown of complex to simpler nitrogenous compounds and so silages may be expected to have a higher nitrogen digestibility than the parent material.

Efficiency of utilization of available nitrogen in the ruminant animal depends upon achieving the optimum balance of the degradative and synthetic activities of the rumen microorganisms. If the degradative processes produce more ammonia than the synthetic activities can utilise, then ammonia accumulates and rumen ammonia concentration rises. Such a situation may arise if food contains a high proportion of simple nitrogenous compounds and where the energy supply to the microorganisms is in deficit. The first of these conditions will apply to silage diets and the second might well do so in view of the processes involved in their production.

The work to be reported here was undertaken to evaluate silages as sources of energy and nitrogen. To this end determinations of energy value, metabolisable energy, rumen concentrations of total and individual volatile fatty acids, rumen pH and ammonia concentrations were made. To allow for the differences in silage composition a number of silages produced in different ways and from different sources was examined.

II

REVIEW OF LITERATURE

A. COMPOSITION OF GRASS

McDonald et al. (1960) gave the figures shown in Table 1 for the composition of a sample of Italian ryegrass.

Table 1

Composition of S22 Ryegrass (g/kg dry matter)

Proximate composition		Carbohydrates		Nitrogenous compounds		Other constituents	
Organic matter	898	Glucose	22	Total nitrogen	30	Lignin	83
Crude protein	187	Fructose	19	Protein	27		
Ether extract	35	Sucrose	35	Non-protein nitrogen	3		
Crude fibre	236	Oligo-saccharides	20				
Nitrogen-free extractives	441	Fructans	56				
		Galactan	11				
		Araban	30				
		Xylan	118				
		Cellulose	262				

The grass had a dry matter of 190 g/kg. The composition of grass will vary depending on certain factors such as species, fertilizer treatment, stage of growth, time of cutting and weather conditions at the time of cutting; but basically, the same components are present in relatively similar amounts.

Glucose, fructose, sucrose and fructan are the main water soluble carbohydrates in grass. Their content is extremely variable depending on the species as shown by Waite and Boyd (1953), the stage of growth (Waite, 1957), the weather (Mackenzie and Wylam, 1957) and the fertilizer application (Waite, 1958). Whittenbury et al. (1967) quoted glucose levels of 16 to 23, sucrose 24 to 84 and fructose 10 to 38 g/kg for Italian ryegrass, while Laidlaw and Reid (1952) gave

levels of 12 to 14, 36 to 74 and 13 to 36 g/kg for perennial ryegrass. Mackenzie and Wylam (1957) reported fructan levels of 40 to 210 g/kg for perennial ryegrass.

The structural carbohydrates are of less importance in ensilage than the water soluble fraction but they are the major components governing the digestibility of the grass. Cellulose is a major constituent of plant cell walls. It consists of long chains of β-1-4 glucose units which form micelles encrusted with lignin, hemicelluloses and silica.

Armstrong et al. (1950), Waite et al. (1964), Jarrige and Minson (1964) showed that cellulose content increased with increasing maturity. The latter workers quoted 140 to 240 g/kg for first cuts of S24 ryegrass from April to June. There was little difference in cellulose content when grass was cut at regular intervals at the same vegetative stage of growth.

Hemicelluloses are heteropolysaccharides made up mainly from pentose sugars, with a small proportion of hexoses and uronic acid units incorporated in the structure. The content of hemicellulose increases with increasing maturity. Jarrige and Minson (1964) gave values varying from 120 to 212 g/kg from April to June for S24 ryegrass.

Lignin is a phenolic compound with a high molecular weight. The lignin fraction is insoluble in alcohol, ether, dilute alkali and sulphuric acid (720 g/l). Jarrige and Minson (1964) and Waite et al. (1964) showed lignin content to increase with advancing maturity. The latter authors gave values of 27 and 73 g/kg for April and June cuts of S23 ryegrass.

The main organic acids present in grass herbage are citric, malic, quinic and succinic. Ferguson (1963) stated that herbage samples commonly contained 50 to 90 g/kg of organic acids calculated as malic acid. Hirst and Ramstad (1957) gave values of 9.0, 0.2, 11.7 and 7.3 g/kg for the quinic, succinic, malic and citric acid contents of perennial ryegrass.

The lipid fraction is made up from fats, waxes, phospholipids, galactolipids and sterols. Grass contains 40 to 80 g/kg of lipid material as determined by solvent extraction. Shorland (1961) demonstrated that galactolipids comprised 0.6 of the lipid fraction of grass. Garton (1960) showed that the unsaturated acids formed 0.89 of the total fatty acids of mixed herbage.

Between 0.7 and 0.9 of the total nitrogen of grass is present as protein; the remainder being mainly in the form of free amino acids, amides and nitrate nitrogen. Wilson and Tilley (1965) found that isolated protein preparations from ryegrass, timothy and cocksfoot contained a similar pattern of amino acids. Ferguson and Terry (1954) reported that about half of the soluble organic nitrogen was in peptide form. Free amino acids, amides, and purine plus betaine and choline nitrogen were respectively 0.15 to 0.20, up to 0.10 and approximately 0.12 of the soluble organic nitrogen. The proportion of soluble organic nitrogen as a fraction of the total nitrogen was less when nitrogen was applied as nitrate rather than ammonium salts or when no application was made (Nowakowski et al., 1965; Nowakowski and Cunningham, 1966). Bathurst (1953) reported that the free amino acid fraction was composed mainly of alanine and serine, but a larger number of amino acids contributed to the peptide fraction. Wilman (1965) showed a maximum of 6.5 g/kg of nitrate nitrogen two weeks after an application of nitro-chalk.

Some of the essential macro elements in grass include nitrogen, sodium, potassium, calcium, phosphorus, magnesium, sulphur, chlorine and the trace elements iron, cobalt, zinc, copper, manganese, molybdenum, iodine and selenium. Silica is also present as an integral constituent of the cellulose matrix. Watson and Nash (1960) reported that the ash content declined with increasing maturity and gave values of 70 to 40 g/kg for timothy, harvested from early June to July.

B. CHEMICAL CHANGES OCCURRING DURING THE ENSILAGE OF GRASS

When grass is ensiled the oxygen entrapped within the mass enables the process of respiration to continue. Sugars are oxidised by plant enzymes to form carbon dioxide and water, and heat is liberated. McDonald *et al.* (1966) have indicated that a loss of sugar of about 4.6 g/kg in the dried herbage by enzymic oxidation would cause an increase in temperature of about 3°C in the mass. Dewar *et al.* (1963) extracted enzymes from perennial ryegrass, Italian ryegrass and cocksfoot, and demonstrated pH and temperature optima of 6, and 30 to 43°C respectively. James (1953) showed acceleration of respiration with temperature to be roughly exponential within the range 0° to 30°C. Wylam (1953) reported that the enzymic hydrolysis of grass sucrose and fructan was extremely rapid if the grass was kept under moist conditions after cutting. Respiration decreased as the dry matter increased.

Russell (1908), working with maize, and Mabbitt (1951), working with timothy, showed that protein was broken down to amino acids and unspecified volatile bases by the action of plant enzymes. Macpherson (1952) showed up to one quarter of grass protein to undergo enzymolysis which slowed markedly at pH 5 but did not cease till pH 4.3.

Gibson *et al.* (1958) discussed the sequence of bacterial changes taking place during ensilage. The fresh herbage quickly consumes the entrapped oxygen (if air is excluded). The aerobic bacteria of the herbage die rapidly and organisms capable of anaerobic growth, members of the Klebsiella group *Streptococcus*, *Leuconostoc*, *Pediococcus*, *Lactobacillus*, *Clostridium* and *Bacillus* begin to multiply.

Soluble carbohydrates are fermented to lactic acid during ensilage by two major groups of bacteria (Whittenbury, *et al.* 1967). The homofermentative lactic acid bacteria produce two moles of lactic acid from one of glucose and the heteroferment-

ative type produce one mole of carbon dioxide, one of ethanol and one of lactic acid per mole of glucose utilized (Wood, 1961). Some of the pathways are shown in Table 2.

Table 2

Main Products of Carbohydrate Fermentation by Lactic Acid Bacteria

Homofermentative:

$C_6H_{12}O_6 + 2ADP + 2P_i \longrightarrow 2CH_3.CHOH.CO_2H + 2ATP$

glucose — lactic acid

$C_6H_{12}O_6 + 2ADP + 2P_i \longrightarrow 2CH_3.CHOH.CO_2H + 2ATP$

fructose — lactic acid

$C_5H_{10}O_5 + 2ADP + 2P_i \longrightarrow CH_3.CHOH.CO_2H + CH_3.CO_2H + 2ATP$

pentose — lactic acid — acetic acid

Heterofermentative:

$C_6H_{12}O_6 + ADP + P_i \longrightarrow CH_3.CHOH.CO_2H + C_2H_5OH + CO_2 + ATP$

glucose — lactic acid — ethanol

$3C_6H_{12}O_6 + H_2O + 2ADP + 2P_i \longrightarrow CH_3.CHOH.CO_2H + 2C_6H_{14}O_6 + CH_3.CO_2H + CO_2 + 2ATP$

fructose — lactic acid — mannitol — acetic acid

$C_5H_{10}O_5 + 2ADP + 2P_i \longrightarrow CH_3.CHOH.CO_2H + CH_3.CO_2H + 2ATP$

pentose — lactic acid — acetic acid

ADP adenosine diphosphate
ATP adenosine triphosphate
P_i inorganic phosphate

The organic acids are also attacked by bacteria; the main pathways are shown in

Table 3.

Table 3

Main Products of Fermentation of Organic Acids by Lactic Acid Bacteria

$C_6H_8O_7 + H_2O + ADP + P_i \longrightarrow 2CH_3.CO_2H + H.CO_2H + CO_2 + ATP$

citric — acetic acid — formic acid

$2C_6H_8O_7 \longrightarrow 2CH_3.CO_2H + CH_3.CO.CHOH.CH_3 + 4CO_2$

citric — acetic acid — acetoin

$2C_6H_8O_7 + H_2O + ADP + P_i \longrightarrow 3CH_3.CO_2H + CH_3.CHOH.CO_2H + 3CO_2 + ATP$

citric — acetic acid — lactic acid

$C_4H_6O_5 \longrightarrow CH_3.CHOH.CO_2H + CO_2$

malic — lactic acid

$2C_4H_6O_5 \longrightarrow CH_3.CO.CHOH.CH_3 + 4CO_2 + 4H$

malic — acetoin

$C_4H_6O_5 + H_2O + ADP + P_i \longrightarrow CH_3.CO_2H + H.CO_2H + CO_2 + 2H + ATP$

malic — acetic acid — formic acid

$C_4H_6O_5 + 2H \longrightarrow C_2H_5OH + H.CO_2H + CO_2$

malic — ethanol — formic acid

Bacterial breakdown of nitrogenous constituents is the result of clostridial activity. This is suppressed in the very acid conditions under which successful silage is made.

The grass detailed earlier (Table 1) was ensiled in four experimental silos (McDonald et al., 1960). The lactic acid fermentation was satisfactory. The dry

matter content of the silages was 190 g/kg and the mean composition is given in Table 4.

Table 4

Composition of Silage (g/kg Dry Matter)

Proximate composition		Carbohydrates		Nitrogenous compounds		Other constituents	
Organic matter	884	Glucose	tr	Total nitrogen	30.4	Lignin	58
Crude protein	190	Fructose	tr	Protein nitrogen	9.6	Lactic acid	88
Ether extract	48	Sucrose	1	Non-protein nitrogen	20.8	Acetic acid	38
Crude fibre	260	Oligo-saccharides	3	Volatile nitrogen	2.1		
Nitrogen-free extractives	387	Fructans	3				
		Galactan	7				
		Araban	15				
		Xylan	82				
		Cellulose	236				

Under less than ideal conditions of ensilage the normal sequence of bacterial actions may be altered (Gibson et al., 1958). An unstable mass is thus produced in which considerable breakdown of the original material has taken place, resulting in a lowered nutritive value. In some cases, a compost like material with a high pH may be produced, which cannot be fed to animals.

Gibson (1965) in his review "Clostridia in Silage" stated that clostridial growth was suppressed by an accumulation of lactic acid and a decrease in the water activity of the plant material.

Gibson et al. (1958, 1961) identified Clostridium butyricum, Cl. welchii and Cl. bifermentans in grass, and Rosenberger, quoted by Gibson (1965), isolated five other types of clostridia from silage. Gibson et al. (1961) identified the

saccharolytic Cl. butyricum, Cl. paraputrificum and Cl. tyrobutyricum in grass and silage. These convert lactic to butyric acid with a consequent rise in pH, which encourages the activity of the proteolytic clostridia, eg. Cl. bifermentans and Cl. sporogenes.

Macpherson and Violante (1966) have shown the presence of ornithine, putrescine and cadaverine in farm silages. They showed an increase in the volatile nitrogen, from 0.032 to 0.182 of the total nitrogen, when the pH increased from 3.55 to 5.05. Bélanger (1964) in a study of volatile bases of grass silage found trimethylamine in levels up to 70 mg/kg of silage.

Kapelle and Postma (1952), cited by Watson and Nash (1960) reported that the activity of butyric acid bacteria was reduced by lowering the moisture content and Wieringa (1958) has shown that the pH tolerance of butyric acid bacteria decreased with increasing osmotic pressure.

Stirling (1951) and Keddie (1954) showed that an increase in dry matter delayed bacterial multiplication. Gouet (1967) confirmed that increased dry matter reduced bacterial population and growth rate, and considered that wilting inhibited spore forming anaerobes, as well as modifying the proportion of different species of gram positive bacteria. Weise (1967) showed a decrease in the ratio of homo to heterofermentative lactic acid bacteria on wilting.

Murdoch (1960), working with wilted lucerne, showed a reduction in volatile acids and bases with increasing dry matter and Gouet et al. (1965) showed a similar effect on lactic acid formation. Macpherson (1952) stated that during wilting there was a rapid accumulation of amides and that 0.16 of the true protein was broken down after a twenty-four hour wilt, and Murdoch et al. (1955) showed that wilting reduced in-silo protein breakdown to 0.15 and 0.19 of the true protein as compared with 0.35 and 0.39 for unwilted materials.

C. THE FATE OF FOOD IN THE RUMINANT

1) The Rumen

Ruminant animals are characterised by the possession of a four compartment stomach which allows extensive pre-gastric microbial fermentation of ingested food. This enables ruminant animals to digest large amounts of cellulose.

The first two compartments of the stomach of the adult ruminant are the reticulum and the rumen (the reticulo-rumen). These comprise about 0.85 of its total capacity as measured by wet stomach contents. The third compartment, the omasum, is smaller, about 0.12 and 0.02 for cattle and sheep respectively (Warner and Flatt, 1965). The omasum leads directly into the glandular abomasum which corresponds to the true stomach in monogastric animals.

Food eaten by ruminants is initially chewed only enough to ensure thorough mixing with saliva to form a bolus of suitable size and consistency for swallowing. This is then carried by muscular contractions of the oesophagus to the reticulo-rumen into which it is ejected with considerable force. The bolus is diluted with copious amounts of saliva, making the dry matter content of the rumen about 100 g/l for sheep (Rogerson, 1958) and 140 g/l for cattle (O'Dell et al., 1963).

In ruminants food undergoes a cyclic process known as rumination which has four distinct phases:-

1) regurgitation of ingesta from the reticulo-rumen,
2) swallowing of the regurgitated liquid,
3) remastication of the solids accompanied by resalivation and,
4) reswallowing of the bolus.

The rechewing during rumination is more thorough than the initial mastication during feeding, and it is at this stage that the greatest physical comminution of the food takes place.

Castle (1950) reported the average rumination time of grazing cattle to be five to six hours, while Schake and Riggs (1966) reported seven to nine for cows kept indoors on various diets. Dukes (1955) indicated fourteen to be the average number of periods of rumination per day varying in duration from a few seconds to more than an hour.

Secondary contractions of the stomach are usually, though not necessarily, associated with eructation of rumen gases (Dziuk and McCauley, 1965). Blaxter and Clapperton (1965) gave average methane production figures of 30 l and 154 l/day for sheep and cattle respectively. Kleiber et al. (1943) and McArthur and Miltimore (1961) reported that carbon dioxide formed 0.65 of the rumen gas and methane 0.27.

Hungate (1966) quoted values of 0.23 to 1.5 days, and 0.35 to 2.3 days for rumen turnover rate for cattle and sheep respectively. The wide variation may be explained by the kind of feed, the level of feeding and the method of measurement. Balch and Campling (1965), working with dairy cows, calculated mean retention times for the reticulo-rumen and omasum to be 60 to 80 hours, while with materials resistant to digestion the time was much longer.

In the rumen the food undergoes breakdown as a result of the activities of the microbial population. The rumen provides an environment which is extremely favourable for the growth of microorganisms. There is a continuous supply of substrate in the form of ingested food, and the end products of fermentation are removed by absorption through the rumen wall and passage to the lower gut. Intimate contact between food particles and microorganisms is achieved by the mixing of the contents of the rumen resulting from the contractions of its muscular walls. The moist conditions of the rumen favour microbial growth and the whole provides an efficient, continuous culture system for anaerobic organisms.

There are two main classes of rumen microorganisms, the bacteria and the ciliate protozoa. Hungate (1966) quoted rumen bacterial counts of 16 to 40

x 10^9 /ml of rumen contents when measured by direct microscopic count, but considerably less, 1 to 8 x 10^9 /ml, when measured by culture counts. The total number of bacteria tend to be higher in animals fed green pasture than those fed dry rations (Gall et al.,1949; Moir, 1951). When a ration was high in concentrates a greater proportion of the bacteria observed by direct count could be cultivated than when hay was the main feed (Maki and Foster, 1957). Bryant and Robinson (1961) found the culture count to be higher just before feeding than one to two hours afterwards.

Rumen organisms may be highly specialised and compete for a few of the available nutrients or have the capacity to utilize many nutrients. As a result the rumen contains a wide diversity of microorganisms. Another suggestion by Hungate (1966) to account for the diversity of the rumen microbial population is based on the concept of "selection for maximum biochemical work".

The rumen microbes include cellulolytic bacteria which produce cellulases capable of hydrolysing cellulose and in some cases cellobiose (Bryant and Burkey, 1953; Hungate, 1950). Some of the commoner species are Bacteroides succinogenes producing acetic and succinic acids, Butyrivibrio fibrisolvens producing butyric, formic and lactic acids and Ruminococcus flavefaciens whose fermentation products are succinic, acetic and formic acids. There are marked differences in the ability of single microorganisms to digest cellulose and hemicellulose from an intact forage (Dehority and Scott, 1967).

The starch and sugar fermenting bacteria include Selenomonas ruminantium yielding acetic and propionic acids as the fermentation products. Streptococcus bovis and Bacteroides amylophilus are both starch digesters; the former producing lactic acid and the latter acetic, formic and succinic acids. The dextran utilising Succinivibrio dextrinosolvens also produces acetic, formic and succinic acids.

Bacteria utilizing lactic acid include Veillonella alcalescens and Peptostreptococcus elsdenii. Both produce acetic and propionic acids and, in addition,

the latter produce butyric, valeric and hexoic acids. Bacteroides ruminicola is one of the most important organisms in proteolysis and deamination of amino acids (Bladen et al.,1961). Smith and Hungate (1958) reported Methanobacterium ruminantium as the chief organism producing methane in the rumen.

Hungate (1966) classified the rumen protozoa into two sub-classes, the holotrichs, which have rows of cilia over their entire body, and the entodiniomorphs, often referred to as the oligotrichs. The holotrichs represented by the genus Isotricha and the genus Dasytricha ferment the soluble carbohydrates and cannot utilize cellulose. The oligotrichs ingest granular starch and can utilize simple and complex carbohydrates including cellulose. The protozoa are strict anaerobes and produce acetic, butyric and lactic acids, carbon dioxide and hydrogen gas. Protozoal counts for sheep and cattle range from 0.6×10^5 to 3×10^6 /ml. Protozoa are sensitive to low pH; Purser and Moir (1959) found that the protozoal population decreased when the pH of the rumen fell to 5.4, and all protozoa are rapidly killed at high acidity.

The material leaving the reticulum enters the omasum by way of the omasal orifice. Balch et al, (1951) suggested that the orifice acted as a valve controlling the movement of ingesta into the omasum. The wet tissue of the omasum constitutes 0.30 and 0.08 parts of the total stomach tissue of mature cattle and sheep (Becker and Arnold,1952; Wardrop and Coombe,1960; cited by Warner and Flatt 1965).

Several workers, Gray et al. (1954), Badawy et al. (1958) and Johnston et al. (1961) have shown that in the omasum 0.33 to 0.64 of the water and 0.40 to 0.71 of the volatile fatty acids are removed from the digesta entering it. The dry matter of omasal contents is relatively constant and higher than that of the reticulum and of the abomasum (Boyne et al., 1956).

The abomasum is the only part of the stomach which secretes digestive juices

and is analagous to the true stomach of monogastric animals. The gastric juice secreted in the fundus region, nearest the omasum, contains pepsin and hydrochloric acid. Fluid entering the abomasum has a pH of 6 (Hill, 1965), and digesta leaving it have a pH between 2 and 3 (Phillipson and Ash, 1965). In the abomasum digesta are mixed with the gastric juice by continuous contractions not correlated with other like movements of the stomach. Badawy et al. (1958) reported a loss of 0.74 of the total volatile fatty acids in the abomasum and Johnston et al. (1961) quoted 0.83.

Sheep and cattle secrete large volumes of alkaline, well buffered saliva, during feed ingestion and rumination. One of the main functions of saliva is the maintenance, within the rumen, of a pH which will favour continuous microbial activity. The rumen contents have a powerful buffering action and may contain between two and five litres of saliva at any one time (Denton, 1957). Rumen liquor contains 57 to 162 m mol/l of total volatile fatty acids (Phillipson, 1942). An acetic acid solution of this strength has a pH of about 3 (Turner and Hodgetts, 1955) but the pH of rumen contents is between 5 and 7.

Saliva is secreted by the salivary glands, which are situated in and around the mouth. In the sheep there are five sets of paired and three unpaired glands (Sisson and Grossman, 1953). The parotid and inferior molar glands secrete continuously, but the others only when feeding (Hungate, 1966), when the saliva will assist in mastication and swallowing. The food bolus may contain up to four times its weight of saliva (Balch, 1958).

The composition of mixed saliva for sheep is shown in Table 5.

Table 5

Composition of Ovine Saliva (McDougall, 1948).

Dry matter g/l	10 -	14
Ash g/l	7 -	9
pH	8.4 -	8.7
Sodium mg/l	3700 -	4620
Potassium mg/l	160 -	460
Phosphorus mg/l	370 -	720
Bicarbonate mg/l	250 -	430

The concentrations of bicarbonate and phosphate anions vary reciprocally (Hungate, 1966), the bicarbonate buffering at pH 6.4 and 10.3, and the phosphate at 2.1, 7.2 and 12.7.

Tribe and Peel (1963) quoted nitrogen levels for sheep saliva of 340 mg/l. Bailey and Balch (1961a) gave urea, non-urea nitrogen and total nitrogen contents for cows of 62.8, 12.9 and 75.7 mg/l respectively.

Daily saliva production for sheep has been quoted as 6 to 16 l (Kay, 1960), 7 to 8 l (McManus, 1961) and 1 to 24 l (Sasaki and Umezu, 1962). Diet affects salivary production as shown in Table 6.

Table 6

Rate of Salivary Secretion in Cows (Bailey, 1959).

Diet	Salivary Flow		Eating Rate
	g/g food	ml/min	g food / min.
Dairy cubes	0.68	243	357
Fresh grass	0.94	266	283
Silage	1.13	250	248
Dried grass	3.25	230	83
Hay	3.63	254	70

The rate of secretion of saliva varies with the dry matter intake and with the moisture content of the diet. Putnam et al. (1966b) found the rate of secretion by steers increased from 33 to 54 l/day when food intake increased from 0.008 to 0.026 of the live weight. Wilson and Tribe (1963) found an increase from 3 to 5 l/day with sheep when the level of intake of dry matter was increased from 600 to 1400 g/day. Stewart and Dougherty (1958) found that water administration into the rumen reduced the rate of flow of saliva. The rate of secretion of saliva is greatest during eating or rumination. Bailey and Balch (1961a) gave the relative secretion rates for cows as 10, 20 and 25 ml/min. for resting, eating and ruminating respectively. The same authors (1961b) considered the rate of secretion during the rest period tended to be greater after small meals than after larger meals.

C. 2) Fate of Carbohydrate in the Rumen.

Carbohydrates are the chief source of energy for both rumen microorganisms and the ruminant animal. Sugars, polysaccharides and heteropolysaccharides are converted in the rumen to microbial cells, carbon dioxide, methane and volatile fatty acids. The latter are waste products of the cells but a source of energy for the host animal.

Carbohydrate breakdown takes place anaerobically under the influence of microbial enzymes and individual carbohydrates are fermented at different rates depending on their solubility and rate of release from the plant (Bailey,1962).

Phillipson and McAnally (1942) infused mono and disaccharides into the rumen of fistulated sheep and showed that glucose, fructose and sucrose were rapidly fermented in the rumen giving rise to lactic acid and volatile fatty acids. Maltose, lactose and galactose were more slowly fermented and no lactic acid was produced. Maltose, glucose, sucrose, raffinose and fructose are rapidly fermented by rumen contents (Quin,1943, cited by Hungate,1966), and Hungate (1966) showed cellobiose, mannose, D-xylose and L-arabinose to be fermented by rumen bacteria. McNaught (1951) showed that 0.90 to 0.96 of the carbon of maltose, incubated with rumen fluid, could be accounted for by the production of lower volatile fatty acids, lactic acid, bacterial protein, bacterial polysaccharide, carbon dioxide and methane. With the pentoses, xylose and arabinose as substrates, only 0.83 to 0.93 of the carbon of the decomposed sugar was accounted for.

Phillipson and McAnally (1942) reported that starch was fermented in the rumen at a slower rate than the simple sugars, with the subsequent prolonged production of volatile fatty acids.

Starch is degraded by α-amylases secreted by rumen microorganisms. Hobson and Macpherson (1952) isolated amylases of differing activity from Clostridium

butyricum and a Streptococcus from the rumen of sheep. The reaction products were maltose, maltotriose and some glucose. Neither enzyme was active against these sugars but did break down maltotetrose. Mould and Thomas (1958) confirmed α-amylase activity of the protozoa, Isotricha and Dasytricha and Bailey (1958) exposed starch to extracts of Epidinium ecaudatum and produced maltose, some glucose and maltotriose. Nasr (1950) demonstrated differing amylase activity in rumen contents from sheep fed different diets. Activity was greater on a flaked maize diet, lower on a diet of hay plus concentrates and lowest on a casein containing diet. Phillipson and Cuthbertson (1956) maintained that the extent of starch breakdown differed for different sources, maize starch was completely broken down in the rumen while potato starch was present in the ileum and caecum four hours after feeding.

Phillipson and Cuthbertson (1956) suggested that cellulose was broken down by a depolymerase in the rumen, yielding small fragments, which were readily broken down by microorganisms, to oligosaccharides and cellobiose.

Conchie (1954) isolated a β-glucosidase preparation from sheep rumen contents. He also suggested that enzymic depolymerisation of cellulose was the initial step in its utilization, followed by a splitting of glycosidic linkages, which would result in the formation of simple sugars which would then be transformed to volatile fatty acids. Casson and Thomas (1955) reported cellulolytic activity in bovine rumen fluid which was nine times more active at six than twentyfour hours after feeding. Stanley and Kesler (1959) and Festenstein (1959) produced cell free cellulolytically active extracts from rumen fluid. Leatherwood (1965) showed cellulases from different species of bacteria might only produce cellobiose while mixed rumen microorganisms produced glucose. Halliwell (1957a) suggested that a mixture of rumen microorganisms was one of the most powerful sources of cellulolytic enzymes. He also suggested (1957b) that rumen protozoa played a minor role

and are effective only in that they ingest bacteria. Hungate (1943, cited by Abou Akkada, 1965) considered that the cellulase present in extracts of Polyplastron multivesiculatum was a truely protozoal product. Satter et al. (1964), using ^{14}C labelled cellulose and an innoculum from cows fed hay plus concentrates, showed acetic, propionic, butyric and valeric acids to be produced.

Gaillard et al. (1965) demonstrated that extracts of mixed rumen bacteria from grass, hay and clover fed animals showed hemicellulase activity, in that all the monosaccharide constituents of grass hemicelluloses were released; galactose and uronic acids more slowly than the pentoses. Howard (1957) was able to demonstrate xylanase activity with an extract of mixed rumen microorganisms. Walker and Hopgood (1961) hydrolysed a largely insoluble preparation of hemicellulose from wheat hay by an extract of sheep rumen microorganisms with the production of xylose, xylobiose, xylotriose, higher oligosaccharides, glucose and arabinose.

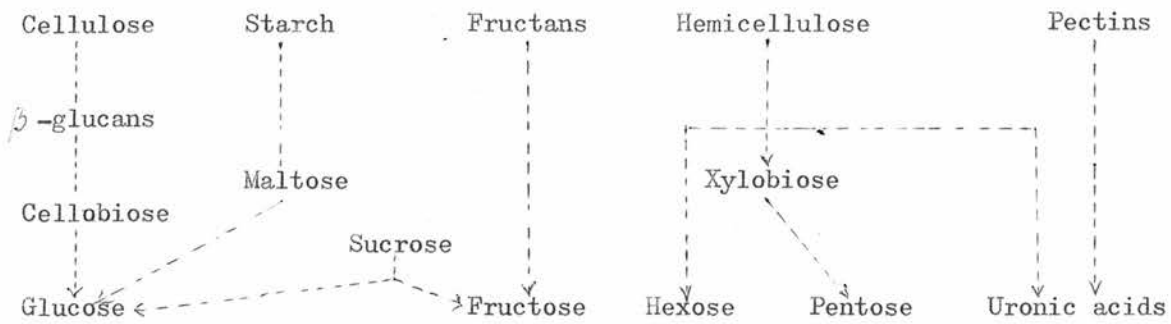
Bailey et al. (1962), working with the rumen protozoa Epidinium ecaudatum, showed it to hydrolyse plant hemicelluloses to their constituent monosaccharides. The mechanism involved initial liberation of arabinose followed by release of xylobiose and finally xylose, glucose, galactose and uronic acids.

Howard (1961), Dehority et al. (1962) reported vigorous in vitro fermentation of pectin by rumen bacteria. Acetic, propionic and butyric acids were the chief products. Abou Akkada (1965) tested cell free extracts of rumen protozoa and confirmed the presence of pectinesterase and a polygalacturonase. The former gave methanol and pectic acid. The polygalacturonic acid was subsequently broken down, by the action of the polygalacturonase on the glycosidic linkages, to give galacturonic acid. However, Howard (1961) claimed only a small proportion of the fermentation of pectins was due to protozoa.

The breakdown of complex carbohydrates in the rumen has been summarised by Halliwell (1961) as shown in Fig. 1.

Fig. 1

Breakdown of Complex Carbohydrates to Simple Sugars



The primary products of breakdown and the simple sugars of the diet are rarely detected in rumen liquor as they are fermented quickly to the short chain volatile fatty acids, to lactic acid and succinic acid. The key intermediate in the breakdown is pyruvic acid which is produced by glycolysis. The stoichiometry of the process is represented in the equation (Walker, 1965):

$$\text{Hexose monophosphate} + 3\text{ADP} + 2\text{P}_i \longrightarrow 2\ \text{Pyruvate} + 3\text{ATP} + 2[2\text{H}]$$

The pathway of fermentation of xylose and other pentoses involves hexose synthesis via the transketolase - transaldolase system and the subsequent degradation of the fructose -1,6- diphosphate or the glyceraldehyde phosphate by the glycolytic pathway to pyruvate as shown in the equation (Walker, 1965):

$$6\ \text{Pentose} + 6\text{ATP} \longrightarrow 4\ \text{Fructose-6-phosphate} + \text{Fructose-1,6-diphosphate} + 6\ \text{ADP}$$

Pyruvate is decarboxylated by at least two distinct mechanisms. Both the coli-aerogenes and the clostridial mechanisms are phosphoroclastic reactions requiring thiamine pyrophosphate (DPT), coenzyme A (CoA.SH) and phosphate and, in addition the latter type require ferredoxin (FD) as hydrogen acceptor (Baldwin, 1965). The mechanisms are shown in Fig. 2.

Fig. 2

Acetate Formation from Pyruvate

Coli-aerogenes phosphoroclastic pathway

$$\underset{\text{Pyruvate}}{CH_3.CO.COO^-} + \underset{\text{Inorganic phosphate}}{PO_4^{III}} \xrightarrow[Mg^{++}\ \ Fe^{++}]{\text{CoA.SH, DPT}} \underset{\text{Formate}}{H.COO^-} + \underset{\text{Acetyl phosphate}}{CH_3.CO.PO_4^{II}}$$

Clostridial phosphoroclastic pathway

$$\underset{\text{Pyruvate}}{CH_3.CO.COO^-} + \underset{\text{Coenzyme A}}{CoA.SH} + FD \xrightarrow[Fe^{++}]{PO_4^{III},\ DPT} CO_2 + \underset{\text{Acetyl coenzyme A}}{CH_3.COS.CoA} + FDH_2$$

$$CH_3.COS.CoA + PO_4^{III} \xrightarrow[\text{Phosphotrans acetylase}]{} CH_3.CO.PO_4^{II} + CoA.SH$$

$$CH_3.CO.PO_4^{II} + ADP \xrightarrow[\text{Acetokinase}]{} CH_3.COO^- + ATP$$

The conversion to acetate and carbon dioxide is an oxidative process and one of the primary pathways of energy metabolism in rumen microorganisms, yielding a high energy bond in the form of acetyl CoA or acetyl phosphate which are interconvertible. The latter can be used to phosphorylate adenosine diphosphate (ADP) as shown,

$$\text{Pyruvate} + CoA.SH + ADP + H_2O \longrightarrow \text{Acetate} + CO_2 + 2\,[H] + ATP$$

There are two pathways known for the conversion of pyruvate to propionate (Baldwin, 1965). These are illustrated in Fig. 3.

Fig. 3

Propionate Formation from Pyruvate

a) Dicarboxylic acid pathway.

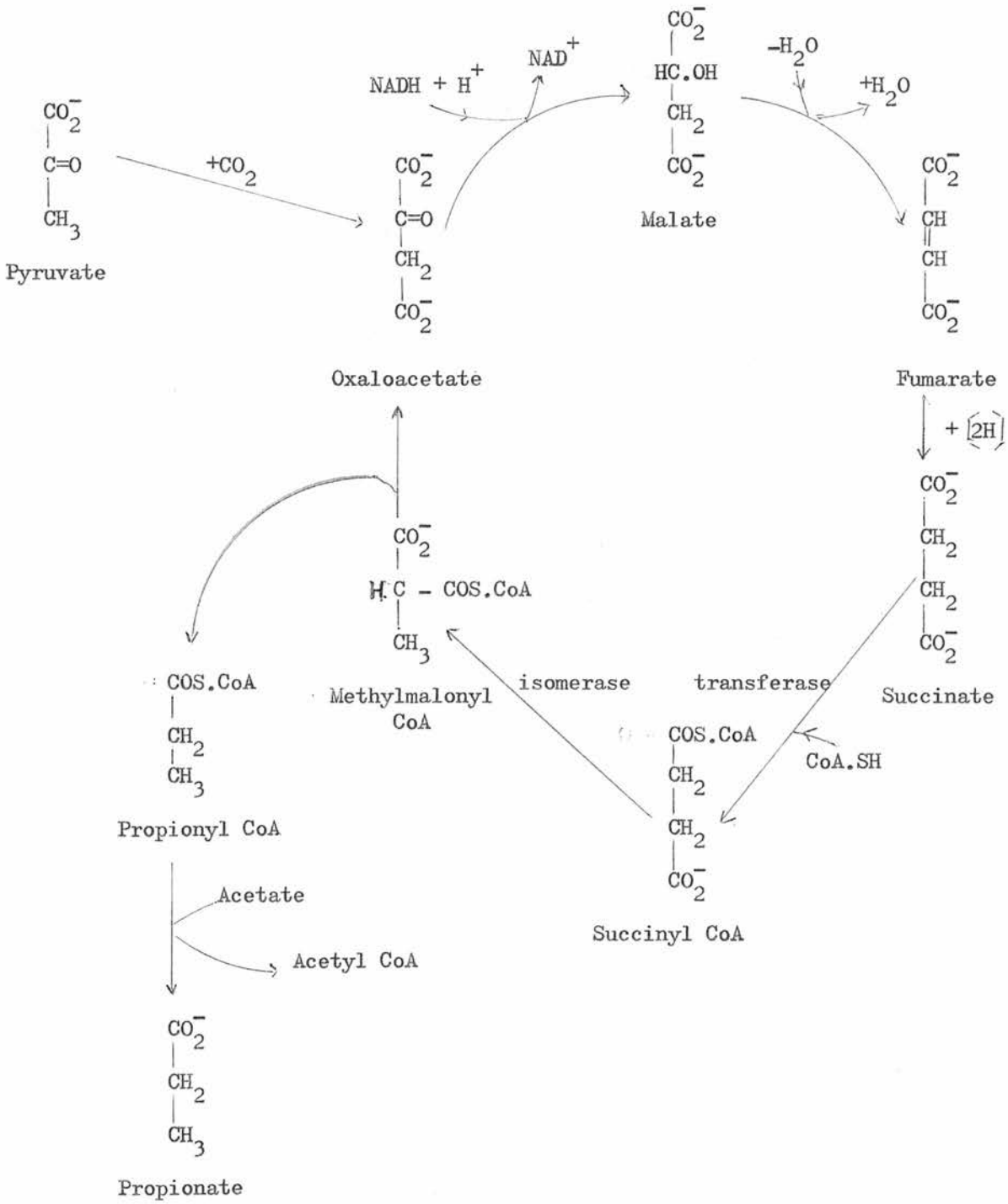
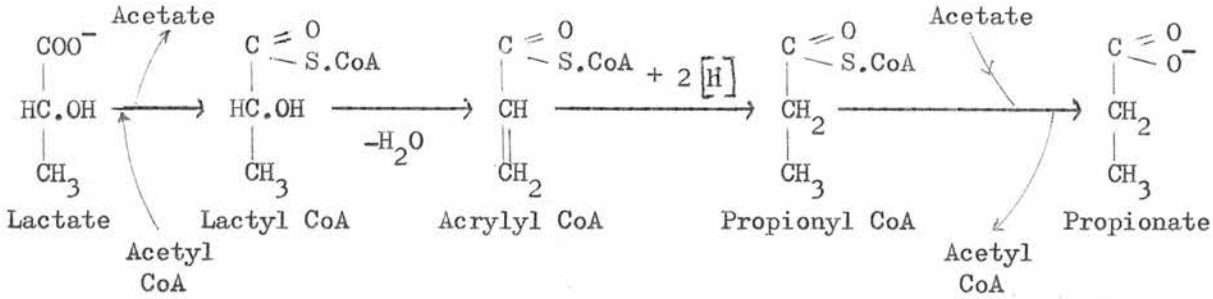


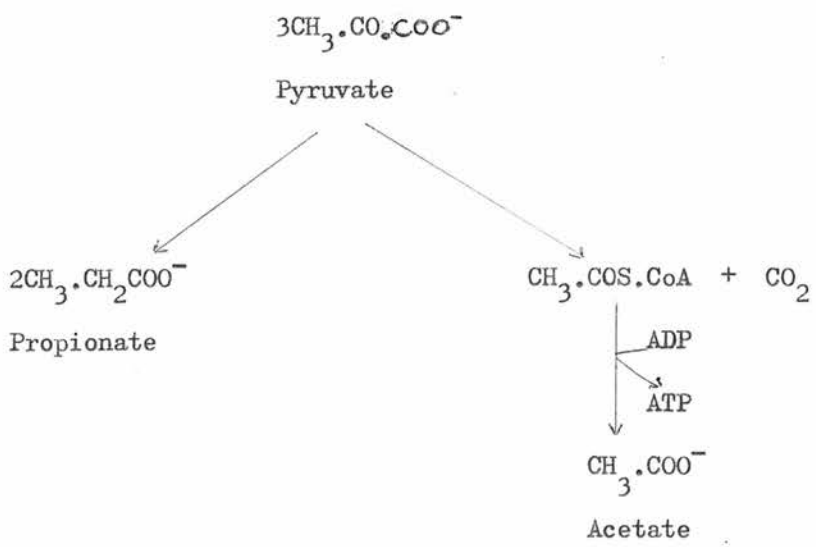
Fig. 3 (Cont'd)

b) Direct reductive pathway.



Blackburn and Hungate (1963) considered that the major part of rumen propionate was produced by the direct oxidative pathway. Baldwin et al. (1962; 1963) demonstrated the existence of both pathways and that 0.7 to 0.9 was formed by the direct reductive pathway and, with increased carbohydrate availability in the diet, the contribution of the propionate produced by the direct reductive pathway increased.

In the production of propionate from pyruvate by the two pathways described, hydrogen, produced by glycolysis and the breakdown of pyruvate, is effectively removed. One mole of acetate is produced for each two moles of propionate as shown:



The net result is removal of hydrogen and the production of one ATP for every three moles of pyruvate metabolised (Stanier et al., 1971).

Baldwin (1965) gave two pathways (Fig. 4) for butyrate synthesis by anaerobic microorganisms. One involving a reversal of β-oxidation, while the alternative involved the formation of malonyl CoA. The pathway via malonyl CoA removes hydrogen but there is no net generation of ATP. The acetoacetyl pathway also removes hydrogen but there is a net generation of one mole of ATP per mole of butyrate formed.

Walker (1965) suggested that where acetate was the source material for butyrate synthesis, the process envolved a net loss of energy:

$$2 \text{ Acetate} + 2\text{ATP} + 2[2\text{H}] \longrightarrow \text{Butyrate} + 2\text{ADP} + 2\text{P}_i$$

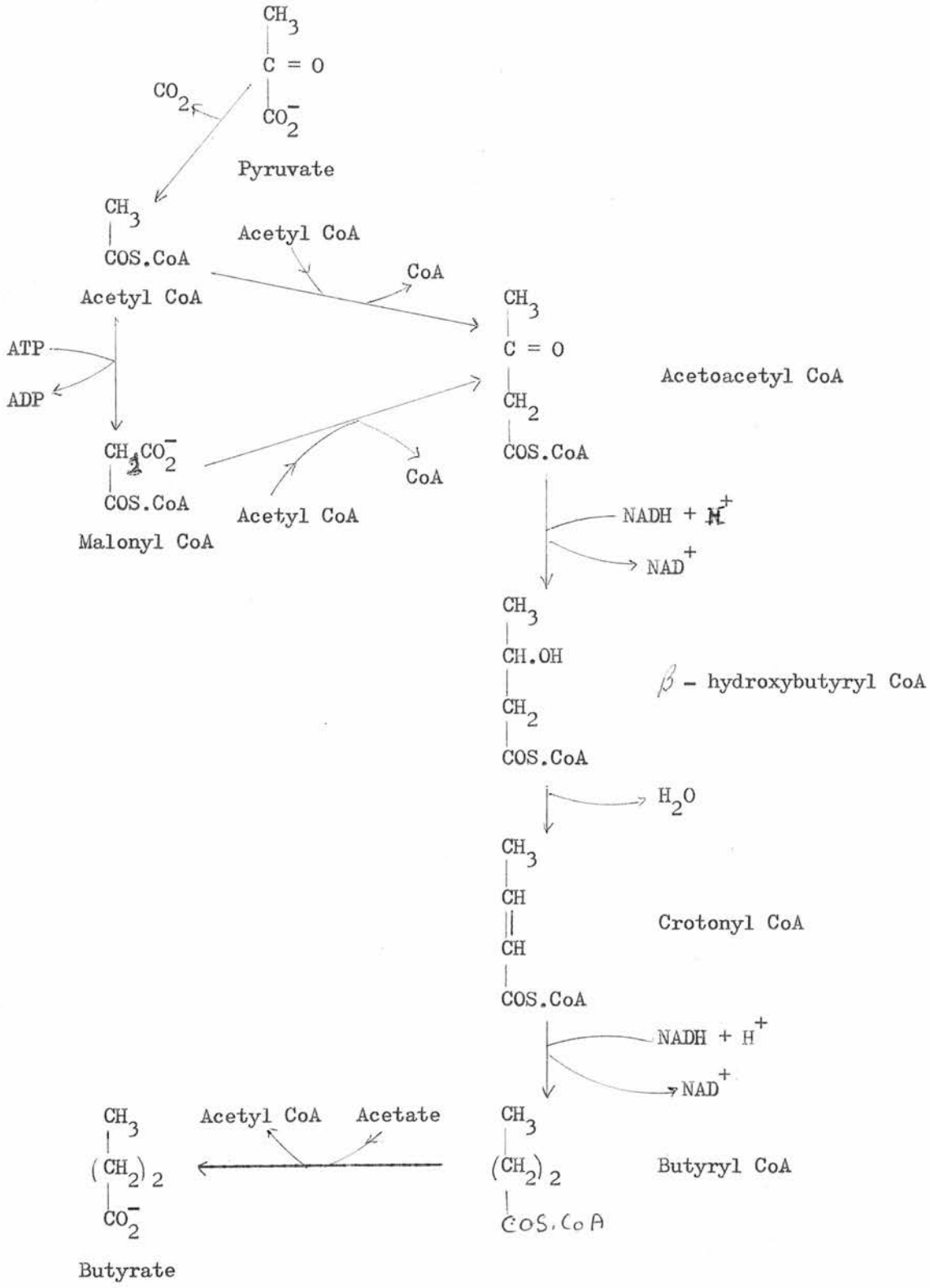
Carbon dioxide in the rumen arises from dietary carbohydrate by oxidative decarboxylation of pyruvic acid and also from the interaction of organic acids with salivary bicarbonate (Hungate, 1966).

The pathway of methane formation is unknown. Methane is synthesised by reduction of carbon dioxide by methanogenic bacteria (Carroll and Hungate, 1955; Bryant, 1965; Williams et al., 1963). The primary precursors in the rumen appear to be formate, carbon dioxide and hydrogen (Hungate, 1966). Carroll and Hungate (1955) found that formate was not itself reduced to methane but supplied the hydrogen to reduce the carbon dioxide. Wolin et al. (1963) have demonstrated synthesis of methane from hydrogen and carbon dioxide by cell free extracts of Micrococcus omeleanskii. The reaction required the presence of ATP and coenzyme A. Ferredoxin stimulated the reaction. The terminal reaction in methanogenesis appeared to envolve the reductive cleavage of methylcobalamine to form methane and cobalamine (Blaylock and Stadtman, 1963).

A large part of the energy derived from the ruminant's food is in the form

Fig. 4

Butyrate Formation from Pyruvate



of the volatile fatty acids. Bergman et al. (1965) quoted 0.8 of the animal's energy expenditure is provided as volatile fatty acids. Leng and Leonard (1965) suggested that volatile fatty acids comprised 0.7 of the apparent digestible energy of the ration.

Halse and Velle (1956) and Gray et al. (1950) using in vitro techniques estimated total daily acid production in sheep of 1.11 mol and 3.4 to 3.9 mol respectively. Carroll and Hungate (1954) and Hungate et al. (1961) using similar methods quoted 23.9 mol and 63.4 mol /day in cattle. Balch (1958b) using combined in vitro and in vivo determinations quoted 25 to 54 mol /day for cattle while Bath et al. (1962) obtained similar values by measuring acid removal from the rumen.

The total daily acid production for sheep fed different diets, measured using a continuous infusion of a single labelled acid or of mixed acids labelled in different ways, is given in Table 7.

Table 7

Daily Production of Volatile Fatty Acids (VFA) in Sheep

Diet	Total VFA (mol/day)	
Grass cubes	5.4	(Bergman et al., 1965)
Lucerne chaff	5.36	(Leng and Leonard, 1965)
Maize (400g) + Lucerne chaff (200g)	5.20	(Leng and Brett, 1966)
" (300g) + " (300g)	4.48	
Wheaten straw (450g) " (50g)	2.42	
Wheaten + Lucerne hay (360g)	3.30	(Gray et al., 1966)
" + " (450g)	4.48	
" + " (650g)	6.42	
Lucerne hay (600g) + Wheaten hay (400g)	4.94 - 4.48	(Weller et al., 1967)
Lucerne hay (500g) + Wheaten hay (500g)	3.94	(Gray et al., 1967)

Continuous infusion techniques offer an opportunity to measure the inter-conversion of the rumen acids. Leng and Brett (1966) reported the extent of propionate interconversion with acetate or butyrate was small. Conversion of

acetate to butyrate accounted for 0.45 of the butyrate produced while the conversion of butyrate to acetate was between 0.06 and 0.13 of the acetate produced in the rumen. Weller et al. (1967) reported that 0.5 to 0.8 of the total butyrate was produced from acetate.

Phillipson and McAnally (1942) postulated that volatile fatty acids were absorbed before they reached the abomasum. Large quantities of acids were produced in the rumen while only small quantities were detected in the abomasum. The addition of sodium acetate or butyrate to the rumen did not increase the level of acid in the abomasum, and volatile fatty acids were not decomposed by rumen microorganisms. Barcroft et al. (1944) found that blood draining the rumen had a higher concentration of volatile fatty acids than did peripheral blood.

Sutton et al. (1963) and Annison (1965) showed that absorption of volatile fatty acids from the rumen was more rapid at pH 5.5 than 7.5. Danielli et al. (1946), Gray (1948) and Sutton et al. (1963) showed that the rates of absorption of the individual acids decreased progressively from butyric to propionic to acetic acid but Weller et al. (1967) using ^{14}C labelled acids showed that the acids disappeared from the rumen at about the same rate.

Annison (1965) could find no evidence for an active transport system across the rumen epithelium. Absorption is apparently a passive uptake in response to a concentration gradient between rumen fluid and blood (Ash and Dobson, 1963).

Pennington (1952) demonstrated that butyrate was metabolised to ketones mainly acetoacetate in its passage through the rumen wall. Annison et al. (1957) found an increase in blood ketones after ruminal infusion of butyrate and Roe et al. (1966) showed these to be mainly β-hydroxybutyrate (BHBA). Acetic and propionic acids pass almost unchanged across the rumen wall into the portal blood where, with the BHBA, they are carried to the liver. In the liver propionic acid is either oxidised or converted to glucose (Annison et al., 1963; Leng and Annison, 1963).

The acetate and BHBA pass almost unchanged, from the liver, into the peripheral circulation, then to the tissues and organs, where they are used as sources of energy and fatty acids (Armstrong, 1965).

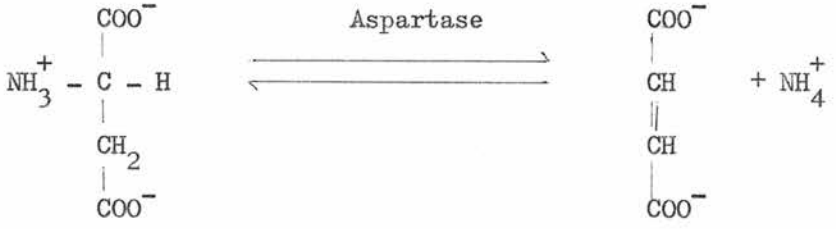
The concentration of volatile fatty acids in the rumen is the result of a balance between the rates of production and absorption, the movement of rumen contents along the digestive tract, the dilution of rumen fluid with saliva, the movement of water across the rumen epithelium and the rate of uptake of volatile fatty acids by the rumen microorganisms. Leng and Brett (1966) gave regression equations to estimate the production of individual volatile fatty acids from their molar concentration in the rumen.

C. 3) The Fate of Nitrogenous Compounds in the Rumen

The nitrogen compounds presented to the rumen microorganisms are chemically diverse. Proteins are the major component of the nitrogen fraction of forages. They also contain non protein nitrogen compounds such as amino acids, peptides, amides, amines, ammonium salts, nitrates and nitrites. Compounds such as biuret and urea may be included in the diet. Non protein nitrogen may form as little as 0.04 of the total nitrogen in soyabean (Krober and Gibbons, 1962) and up to 0.7 in unwilted silages (McDonald et al., 1966b). Hughes (1967) reported the free amino acid content of some silages could be as high as 0.2 and 0.3 of the total nitrogen. The total nitrogen content of rumen contents is between 3 and 5 g/l. The non protein nitrogen fraction is made up from 100 to 600 mg/l ammonia nitrogen, 5 to 100 mg/l amino nitrogen and 2 to 50 mg/l peptide nitrogen (Lewis, 1961).

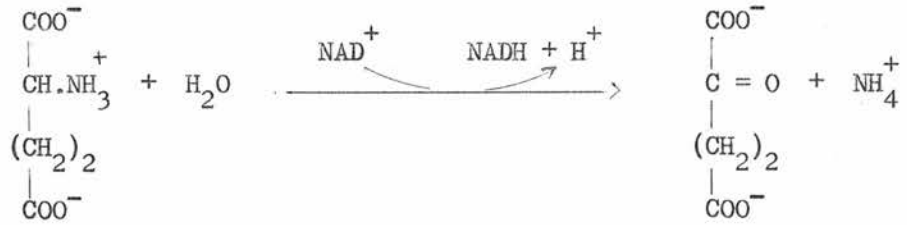
Proteins entering the rumen are broken down to peptides and amino acids. Weller et al. (1958) fed wheaten hay to sheep and found that the protein disappeared completely in sixteen hours. Borchers et al. (1965) incubated strained rumen liquor with a variety of foods and showed considerable variation in the quantities of amino acids produced. Proteolytic activity in the rumen is vested in the micro-organisms, and Blackburn (1965) listed Bacteroides, Selenomonas and Butyrivibrio species among the proteolytic bacteria occurring in the rumen. Among the rumen protozoa Entodinium was shown to have proteolytic activity (Abou Akkada and Howard, 1962) and Blackburn and Hobson (1960) showed the oligotrich protozoa ingested and digested stained casein particles. However, Blackburn and Hobson (1962) considered only 0.1 of total rumen isolates had proteolytic activity. Henderickx and Martin (1963) related protein solubility to their susceptibility to proteolysis, but Blackburn (1965) considered there was little evidence to support such a relation-ship.

Dietary amino acids and those produced by breakdown of dietary protein undergo decomposition by rumen microorganisms to give carbon dioxide and ammonia. Lewis (1955) showed that different amino acids yielded different quantities of ammonia when introduced into the rumen. The ammonia may be produced by non oxidative deamination, under the influence of specific deaminases, as shown in the equation:



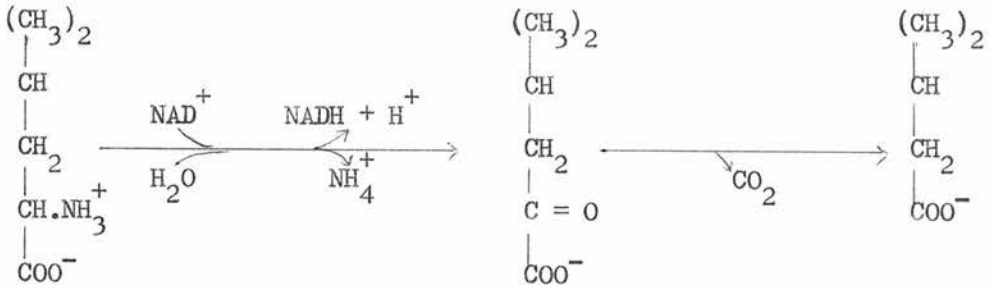
L-aspartate Fumarate

Other amino acids may undergo oxidative deamination as in the case of L-glutamate:



L-glutamate α-keto glutarate

El Shazly (1952) identified several low molecular weight acids in rumen contents, notably iso-butyric, iso-valeric, and 2-methylbutyric, and many workers have confirmed their presence (Annison, 1954; Rhodes and Woods, 1962; Woods and Luther, 1962; Omar et al., 1964; Bath and Rook, 1965). These acids could be produced by oxidative deamination of individual acids as shown for leucine:



leucine ∝-keto iso-caproic acid iso-valeric acid

Iso-butyric and 2-methylbutyric acids are similarly produced from valine and iso-leucine. δ-amino valeric acid was converted by clostridia to n-valeric acid, propionic and acetic acids, and ammonia.

$$2H_3^+N.(CH_2)_3.\overset{*}{C}H_2.COO^- + 2H_2O \longrightarrow 2NH_4^+ + CH_3(CH_2)_2.\overset{*}{C}H_2COO^- + CH_3.CH_2COO^- + \overset{*}{C}H_3.COO^-$$

valeric acid — propionic acid — acetic acid

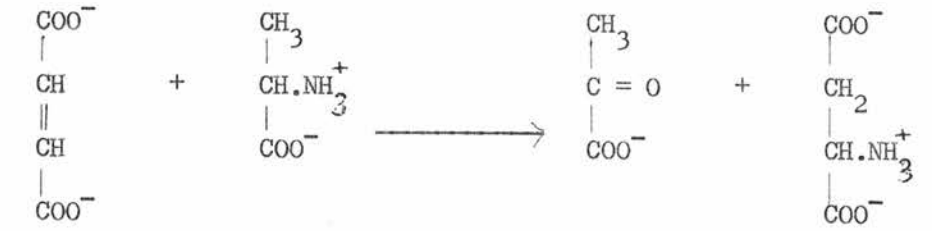
Evidence from the use of labelled δ-amino valeric acid indicated that the fermentation involved reductive deamination followed by β-oxidation of a C_5 to acetic and propionic acids (Barker, 1961).

When L-lysine was incubated with mixed bovine contents, butyric and acetic acids, and ammonia were produced:

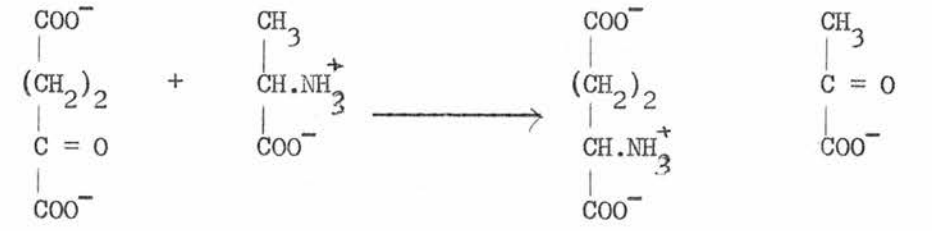
$$CH_2.NH_3^+CH_2.(CH_2)_2.CH.NH_3^+.COO^- + 2H_2O \longrightarrow 2NH_4^+ + CH_3.COO^- + CH_3.CH_2.CH_2.COO^-$$

Evidence from incubation of lysine - 6 - C^{14} with Clostridium Sticklandii indicated cleavage of the lysine between carbon atoms two and three, or four and five (Barker, 1961).

Most amino acids first undergo a process of transamination by reaction with fumaric or ∝-ketoglutaric acid when aspartic and glutamic acids are produced along with the ∝-keto acid:



fumarate alanine pyruvate aspartate



α-ketoglutarate alanine glutamate pyruvate

The amino acids are then deaminated by specific deaminases to give ammonia and the keto acid, which may be used in further transaminations. There is little information on deamination by pure cultures of rumen bacteria. Bladen et al. (1961) found Bacteroides ruminicola was the only species which produced significant amounts of ammonia, but Lewis and Elsden (1955) showed that Selenomonas ruminantium and Peptostreptococcus elsdenii deaminated L-threonine, L-serine and L-cysteine. Warner (1956) demonstrated that protozoa were able to deaminate amino acids.

Lewis (1951 a & b) showed that nitrate introduced into the rumen of the sheep was reduced to ammonia and that nitrate was reduced to ammonia by rumen micro-organisms in vitro; formate, succinate, lactate, citrate, glucose, malate and mannitol were classified as hydrogen donors for nitrate reduction. Wolin et al. (1961) isolated Vibrio succinogenes from bovine rumen fluid and showed it to reduce nitrate to ammonia. Ammonia is also produced in the rumen from guanine, hypoxanthine, xanthine, uric acid, uracil and thymine (Jurtschuk and Hueter, 1955).

Pearson and Smith (1943) showed urea was rapidly hydrolysed to ammonia and

carbon dioxide in the bovine rumen. Jones et al. (1964) showed that intracellular bacterial urease was responsible for ureolysis by rumen contents. They also found that 0.33 of the bacteria isolated from the rumen of a sheep fed hay, barley and urea produced urease. Abou Akkada and Howard (1962) found very little, if any, urease activity in protozoal suspensions.

Microbial protein synthesis has been demonstrated in vitro by a decrease in non protein nitrogen and an increase in microbial protein (Pearson and Smith, 1943; Smith and Baker, 1944). Blackburn (1965) cited work using labelled ^{35}S which showed an increase in bacterial protein of 0.44 after six hours incubation of soya gluten and arabinose. Hungate (1966) estimated that about 10g of microbial protein were synthesised for each 100g of organic matter fermented.

Certain carbohydrate sources enhanced or depressed the conversion of nitrogen to bacterial protein (Houpt, 1958; Mills et al., 1944; Reis and Reid, 1959). Warner (1956) noted that starch or other polysaccharides reduced ammonia concentration and suggested that this was not due to reduced proteolysis or deamination but to increased utilization of ammonia for protein synthesis. Ely et al. (1967) reported 0.3 of the dietary zein was converted to microbial protein when the diet contained a high starch to cellulose ratio but only 0.26 when the ratio was reversed.

McDonald (1954) and McDonald and Hall (1957) showed 0.4 of zein nitrogen and 0.9 of casein nitrogen was converted to microbial protein in the rumen and Weller et al. (1958) showed that rumen microbial nitrogen increased from 0.63 to 0.82 of the total nitrogen after feeding wheaten hay to sheep. Ulbrich and Scholz (1966) showed labelled ^{15}N was rapidly incorporated in rumen microbial bacteria and protozoa. A maximum of about 0.17 of the label appeared in the bacteria of the cattle on the fifth day and 0.13 in the rumen protozoa on the sixth day.

Ruminococcus flavefaciens, R. albus and Bacteroides succinogenes have all been shown to utilize ammonia for cell synthesis (Blackburn, 1965). Bryant and

Robinson (1962), working with isolated strains, found that 0.25 required ammonia only, 0.06 required amino acids, and 0.56 grew with either ammonia or amino acids. Ammonia is used in preference to preformed amino acids by the majority of the proteolytic isolates from a number of genera (Abou Akkada and Blackburn, 1963).

Ammonia absorption from the rumen was first demonstrated by McDonald (1948). Lewis (1957) showed that the portal blood increased as a curvilinear function of rumen ammonia concentration. Gärtner (1963) found greater absorption at higher rumen ammonia levels and concluded that the process was one of simple diffusion. Annison (1965) agreed with this finding and could see no evidence for a mechanism other than simple diffusion. Hogan (1961) showed that ammonia was absorbed more rapidly than the ammonium ion and absorption was more rapid at pH 6.5 than at 4.5. This agrees with the findings of Coombe et al. (1960) who found signs of ammonia toxicity when the pH rose to 7.3.

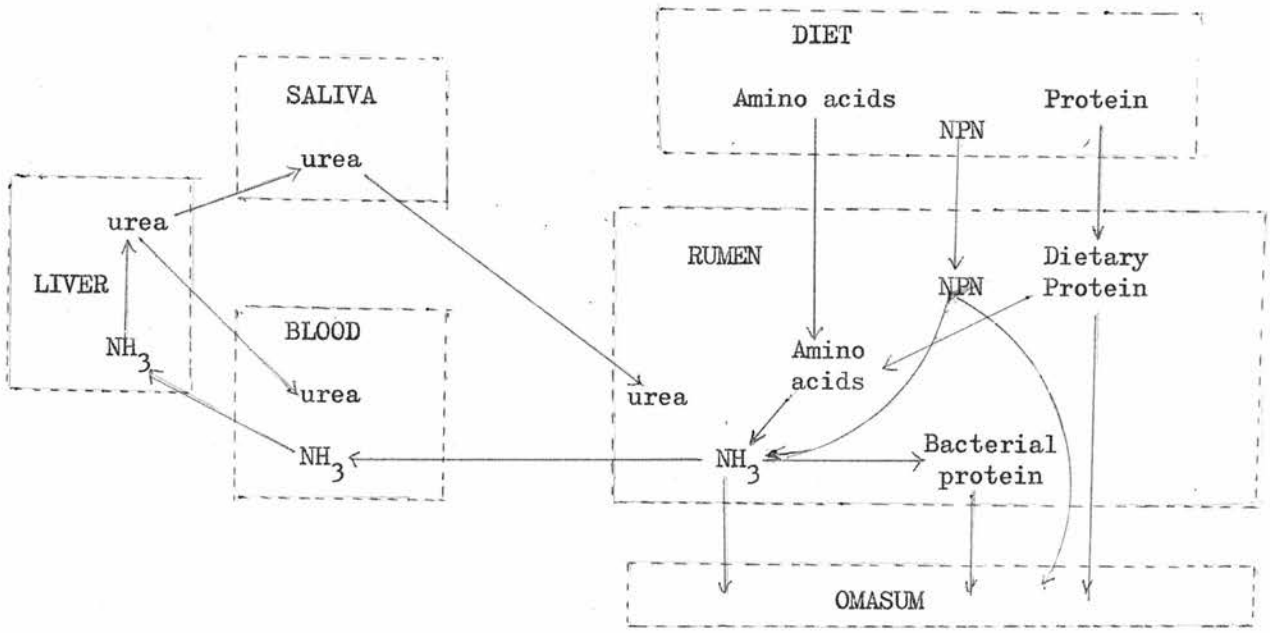
Estimates of the amount of ammonia nitrogen absorbed from the rumen of a sheep per day was four to five grams (McDonald, 1948), but up to fourteen grams has also been reported (Lewis et al., 1957).

Ammonia is converted to urea in the liver. Some urea from the liver returns to the rumen as salivary urea and some passes from the blood across the rumen wall (Le Bars, 1967). The total endogenous influx into the rumen is mainly a function of the blood urea concentration. The amount of urea transferred across the rumen mucosa can be four to six times that of parotid secretion (Cocimano and Leng, 1966).

A scheme showing the fate of dietary nitrogenous compounds in the ruminant is shown in Fig. 5.

Fig. 5

Fate of Dietary Nitrogenous Compounds in the Ruminant



The rumen ammonia pool exists as a balance between the ammonia formed from dietary protein and non protein nitrogen, plus that from endogenous urea, and the ammonia absorbed, and the ammonia utilized by the rumen bacteria.

Tagari et al. (1962) with soyabean meal, Whitelaw et al. (1961) with groundnut meal and Chalmers et al. (1954) with casein reported increased nitrogen retention with heat treatment of dietary protein. The latter workers correlated the improvement in retention with less ammonia production in the rumen. Chalmers and Marshall (1964) showed maximum values of 400 mg/l at three to four hours and 260 mg/l of ammonia nitrogen at one hour post feeding for groundnut and herring meal respectively. They correlated the lower ammonia levels of the herring meal diet with improved utilization as shown by nitrogen balance and an increase in daily milk yield of 0.27 kg for dairy cows. Moir and Somers (1957) for a single ration fed in different ways reported increased retention of nitrogen with more frequent feeding of the diet and correlated this with a decrease in the peak ruminal ammonia levels.

Oyaert and Bouckaert (1960) suggested that at concentrations of ruminal ammonia of about 80 mg/l ammonia nitrogen, there was no loss of nitrogen from the rumen while at a concentration of 130 mg/l there was no loss of protein nitrogen.

C. 4) The Fate of Lipids in the Rumen

Lipids generally compose a small fraction of the dry matter of most ruminant diets but the daily intake can be as high as five hundred grams (Garton, 1961).

Rumen microorganisms hydrolyse glycerides to fatty acids and galacto-glycerides to sugars, the glycerol produced by the hydrolysis is fermented to propionic acid, and the sugars to volatile fatty acids. The unsaturated fatty acids are hydrogenated by the bacteria of the rumen. In vitro work has shown that strict anaerobic conditions are necessary for hydrogenation. Two systems appear to be involved with linoleic acid, one to convert to the mono unsaturated acid, and the second for its complete saturation to stearic acid (Garton, 1965). Abou Akkada (1965) cited work which showed that protozoa were also active in the hydrogenation of fatty acids.

D. EFFECT OF DIET ON RUMEN FERMENTATION

1) pH

The pH of the rumen is affected by the diet and by the proportions of roughage to concentrate in mixed diets. Raun et al. (1962) found a decrease in the rumen pH from 6.5 to 6.3 when the roughage inclusion in the diet was reduced from 0.5 to 0.2 parts. Luther and Trenkle (1967) reported rumen pH values in lambs of 6.5 and 6.4 when the roughage inclusion was 0.6 parts of the diet, which was given in two different physical forms: the pH decreased to 6.2 for both forms when the roughage inclusion was 0.4

When a diet containing 0.45 parts of roughage was replaced by one with 0.08 parts of roughage, the mean rumen pH of cows decreased from 6.25 to 5.73 (Balch and Rowland, 1957). Emery and Brown (1961) found when the roughage portion of the diet was reduced from 0.6 to 0.07 the rumen pH of cows at three hours post feeding declined from 6.3 to 5.8 but the grain in the latter diet was dehydrated and pelleted.

Chou and Walker (1964) reported pH values in the rumen of sheep of 7.67 and 6.43 when a lucerne diet was completely replaced by wheat. Briggs et al. (1957) and Eadie et al. (1967) confirmed the effect of increasing dietary proportions of concentrate in causing a decrease in ruminal pH.

In comparing the effect on rumen pH of the physical form of all roughage diets, Cullison (1961), with steers fed long and pelleted hay, reported pH values of 6.28 and 5.22 respectively for the rumen contents sampled between one and three hours after feeding. Kromann and Meyer (1964) reported a decrease in ruminal pH from 6.70 to 6.55 when a pelleted replaced a chopped hay ration. Rumen pH values of 6.6 and 6.5 were reported when lambs were given ground and pelleted hay respectively (Luther and Trenkle, 1967).

The physical form of mixed roughage plus concentrate diets and ruminal pH was investigated by Balch and Rowland (1957) who reported mean pH values of 6.20 and 6.03 for long and ground hays respectively in the mixed diet. Woods and Luther (1962) found the rumen pH values of sheep four hours after feeding to be 6.4 and 5.9 for a ration of long hay plus concentrate, and the same diet pelleted. For fistulated lambs Rhodes and Woods (1962) reported mean pH values at four hours post-feeding of 5.85, 5.90, 5.89 and 5.96 for mixed diets of finely ground corn, coarsely ground corn, pelleted finely ground corn and pelleted coarsely ground corn, respectively.

The decrease in rumen pH resulting from the pelleting of a mixed concentrate roughage diet may be accentuated when the concentrate inclusion is high. Luther and Trenkle (1967) reported rumen pH values of 6.7, 6.5, 6.4 and 6.7 for diets containing 0.2 parts of concentrate given ground, ground and pelleted, roughage portion pelleted and with both roughage and concentrate pelleted. For diets in the same physical form but containing 0.8 parts of concentrate the figures were 6.3, 5.8, 6.2 and 5.9. Putnam et al. (1966) with steers fed unpelleted and pelleted coarsely ground mixed diets of 0.10 and 0.75 parts of concentrate reported rumen pH values of 6.7 and 6.6 respectively for the low concentrate inclusion and 6.3 and 6.1 respectively for the high concentrate inclusion. However, Bull et al. (1965) did not find a similar effect on rumen pH; with 0.5 parts of concentrate the rumen pH values were 6.5 and 5.7 for the long and pelleted forms and with 0.8 parts, 6.1 and 5.7. Kromann and Meyer (1964) reported a decrease in rumen pH from 6.1 to 5.95 when a diet of 0.8 parts of barley was pelleted.

Christian and Williams (1957) fed fresh and dried grass to sheep and reported rumen pH values at pre-feeding of 7.15 and 6.7 respectively and minimum values of 6.7 and 6.6 respectively. Hinders et al. (1961) and Ghorban et al. (1966) fed alfalfa hay and dehydrated alfalfa pellets to cattle. The former workers gave rumen pH

values of 6.9 and 6.0 respectively, while the latter quoted 7.1 and 6.4. Mean rumen pH values in cattle of 6.7 and 6.3 were reported with diets of flaked corn plus silage and hay, and cracked corn plus silage and hay (Ghorban et al., 1966). Davis and Stallcup (1967) reported slightly decreased rumen pH values when raw soya-bean was replaced by the meal, but Chou and Walker (1964) found little change in pH with the cooking of a potato diet.

D. 2) Total Volatile Fatty Acids

Weston and Hogan (1967) with all roughage diets reported increased concentration of total volatile fatty acids (TVFA) in the rumen from 113 to 130 m mol/l when chopped hay was replaced by finely ground material. The same authors could find no difference when a similar experiment was carried out using wheaten hay instead of lucerne. Mahapatro and Leffel (1964) found no variation in total acid concentration between long, coarsely ground and finely ground hay. Wright et al. (1963), with sixty lambs on an all roughage diet in different forms, found the TVFA concentration to decrease from 215 for pellets to 196 for crushed pellets, 189 for finely ground, 173 for coarsely ground and 165 m mol/l for long hay.

With mixed diets of roughage and concentrate, Woods and Luther (1962) found maximum TVFA concentration in the rumen of sheep of 93 m mol/l when the whole ration was pelleted; 64 m mol/l when the hay was long; and increased levels of 75 and 88 m mol/l when part of the diet was pelleted. Clanton and Woods (1966), Putnam et al. (1966) working with steers, and Jorgensen and Schultz (1963) with dairy cows, reported similar increases with pelleting. Luther and Trenkle (1967) showed little difference from 78 to 81 m mol/l in TVFA concentration in the rumen of lambs when fed ground and pelleted diets of roughage and concentrate in the ratio of 4 to 1, and 64 to 70 m mol/l when the ratio was 1 to 4.

The effect of the fibre content of the dietary roughage on the TVFA concentration in the rumen is somewhat confusing. Bath and Rook (1961) reported a range of only 114 to 134 m mol/l in TVFA concentration in the rumen contents of two cows fed indoors with S23 perennial ryegrass at eight stages of growth. Bath and Rook (1965) quoted rumen TVFA concentrations of 122 and 92 m mol/l for diets of Italian ryegrass cut in March and May respectively. They also quoted 112 and 103 m mol/l for April cuts of perennial ryegrass and timothy / meadow fescue respectively and

93 and 114 m mol/l for May cuts of cocksfoot and lucerne respectively. The TVFA content of rumen liquor from two fistulated New Zealand sheep at pasture ranged from 101 to 187 m mol/l, but there was no definite pattern of change throughout the year (Johns, 1955).

Williams and Christian (1959) fed twelve silages of varying organic matter digestibilities and reported TVFA values in the rumen of sheep to range from 49 to 87 m mol/l two hours after feeding. There did not appear to be a correlation between organic matter digestibility and the level of TVFA in the rumen. A late cut grass silage diet gave a level of 124 m mol/l of TVFA in the rumen of cows and an early cut silage with a low fibre content gave 87.5 m mol/l, while hay, with a fibre content similar to that of the late cut silage, gave a concentration in the rumen of TVFA of 95 m mol/l (Bath and Rook, 1965).

Briggs et al. (1957) reported higher maximum TVFA concentrations of 111 to 165 m mol/l in the rumen of sheep fed a diet of lucerne compared with 64 to 107 m mol/l for a diet of wheaten plus lucerne chaff, while replacement of part of each of the roughages by maize, wheat, oats or starch resulted in higher acid concentrations. Raun et al. (1962) reported a slight decrease in TVFA level from 89 to 70 m mol/l when the proportion of concentrate in the diet of sheep was increased from 0.5 to 0.8 parts. Luther and Trenkle (1967) showed little change in TVFA concentration when the concentrate inclusion in the diet of lambs was 0.2, 0.4 and 0.6 parts. Balch and Rowland (1957) reported mean values of 132 and 142 m mol/l of TVFA in rumen liquor samples taken at hourly intervals from dairy cows on a hay and concentrate diet when the concentrate was increased from 0.56 to 0.92. With the increase in the proportion of concentrate, a wider range of acid values was observed, from 106 to 161 with the low concentrate inclusion, and 67 to 166 m mol/l with the low hay inclusion.

Eusebio et al. (1959) fed diets of alfalfa hay (0.69) and corn meal (0.31)

at two levels of intake, and reported TVFA values of 103.8 m mol/l at 8.3 kg/day, and 140.3 m mol/l at 8.7 kg/day. Corn meal fed at 5.4 kg/day gave values of 125.6 m mol/l.

Working with steers, Putnam et al. (1966) found the rumen TVFA concentration increased from 77 to 97 m mol/l by decreasing the proportion of hay from 0.9 to 0.25. Templeton and Dyer (1967), using mean TVFA values of rumen contents sampled pre and post feeding, reported an increase from 107 to 137 m mol/l when 0.5 of an all hay diet was replaced by concentrate. Clanton and Woods (1966) did not find any increase in TVFA concentration in the rumen sampled at three hours post feeding when the hay was replaced by 0.25 and 0.5 parts of concentrate but these diets were pelleted. The inclusion of concentrate at 0.75 parts of the diet showed a decrease in rumen TVFA concentration from 109 to 98 m mol/l. This was confirmed by Templeton and Dyer (1967) when the concentrate part of their ration was 0.8 parts.

El Shazly (1952) noted that heat treatment of the food increased the level of ruminal TVFA. When dried grass replaced frozen grass as the sole food the prefeeding TVFA concentration increased from 72 to 96 m mol/l and the post feeding levels from 127 to 142 m mol/l. Christian and Williams (1957) found that heat treatment of the grass diet increased prefeeding levels of rumen acids from 52 to 65 m mol/l, but decreased the rise following feeding.

When cracked corn replaced pelleted corn in the diet of steers receiving pelleted alfalfa, Clanton and Woods (1966) found a reduction from 117 to 98 m mol/l in ruminal TVFA concentration at three hours after feeding. A similar fall was obtained with ground or pelleted alfalfa. Eusebio et al. (1959) replaced an all corn meal diet with flaked corn and found no difference in rumen TVFA concentration.

D. 3) Individual Volatile Fatty Acids

Balch and Rowland (1957) fed a mixed concentrate roughage diet and reported a range of 0.656 to 0.595, 0.213 to 0.183 and 0.149 to 0.118 for the molar proportions of acetic, propionic and butyric acids respectively when the hay was in the long form, and 0.546 to 0.465, 0.353 to 0.273 and 0.142 to 0.090 with the ground hay. The mean values were 0.617 and 0.507 for acetic acid, 0.188 and 0.313 for propionic acid and 0.112 and 0.118 for butyric acid for the long and ground hay respectively.

The molar proportions of acetic acid in the rumen of lambs, on an all roughage diet, sampled at four hours post-feeding, were 0.625 on long, 0.568 with coarsely ground and 0.475 with finely ground hay. The proportions of propionic and butyric acids were 0.238 and 0.108 on long, 0.271 and 0.136 with coarsely ground and 0.285 and 0.239 with finely ground hay (Wright et al., 1963). Hogan and Weston (1967) reported an increase from 0.627 to 0.645 in the molar proportion of acetic acid in the rumen of sheep sampled at 30. 90 and 150 minutes after feeding when a lucerne diet was ground.

Luther and Trenkle (1967) compared a ground roughage diet with the same diet pelleted, and found pelleting decreased the molar proportion of acetic acid in the rumen contents of lambs, from 0.695 to 0.662, increased propionic acid from 0.209 to 0.221 and of butyric acid from 0.083 to 0.090. Jorgensen and Schultz (1963) also found decreases in acetic acid from 0.605 to 0.575 and increases in propionic acid from 0.20 to 0.22 and little change in butyric acid proportions in the rumen when the concentrate part of a mixed diet was pelleted, but when the roughage portion was pelleted there was a decrease in the butyric acid proportion from 0.170 to 0.155 and a large increase from 0.20 to 0.27 in propionic acid in the rumen. Clanton and Woods (1966) reported similar results in the rumen contents of steers sampled at three hours post-feeding when the roughage (0.25) part of the ration was

pelleted and the concentrate was pelleted corn. However, when the concentrate was cracked corn, pelleting of the roughage increased the molar proportion of acetic acid from 0.501 to 0.550, and decreased that of propionic acid from 0.310 to 0.285, and butyric acid from 0.126 to 0.111.

Pelleting a diet of 0.8 parts of concentrate decreased the molar proportion of acetic acid in the rumen of lambs from 0.439 to 0.395 and propionic from 0.429 to 0.403, and increased the proportion of butyric acid from 0.096 to 0.154 for the ground and pelleted diets respectively. Pelleting a diet of 0.2 or 0.4 parts of concentrate did not affect the distribution of the volatile fatty acids in the rumen (Luther and Trenkle, 1967).

Ensor et al. (1959) reported the molar proportions of acetic acid of 0.705 and 0.679, and propionic of 0.202 and 0.195, and butyric of 0.075 and 0.087 in the rumen of cows on long and pelleted hay diets respectively.

Woods and Luther (1962) reported the molar proportions of acetic and propionic acids in the rumen of sheep sampled at three hours post-feeding on diets of long hay and concentrate were 0.53 and 0.32 respectively while with a pelleted diet they were 0.31 and 0.44 respectively. Ensor et al. (1959) confirmed the change in acid distribution when a mixed concentrate roughage diet was pelleted but found the proportion of butyric acid decreased from 0.156 to 0.058 when the ration was pelleted.

Bath and Rook (1965) studied the effect of fibre content of the diet on the molar distribution of the major volatile fatty acids of the rumen. S23 ryegrass was fed at six stages of growth from leafy, at the beginning of April, to mature at the end of May. For two cows, over a twelve hour sampling period, the mean values ranged from 0.585 to 0.658, 0.240 to 0.187 and 0.139 to 0.116 for the molar proportions of acetic, propionic and butyric acids respectively, but there did not appear to be any effect of stage of maturity on the pattern of acid proportions in the

rumen. Parks et al. (1964) fed ryegrasses with cellulose contents varying from 268 to 425 g/kg and sampled rumen contents at four hours after feeding. They found that the molar proportions of acetic acid increased from 0.60 to 0.80, while those of propionic and butyric acids decreased from 0.29 to 0.15, and 0.13 to 0.04 respectively. Armstrong (1964) fed diets of dried S23, S24, S37 and timothy to sheep. There were differences between species in the molar proportions of acetic and propionic but not of butyric acid.

The molar proportions of the major volatile fatty acids in the rumen of dairy cows sampled at four hours after feeding were 0.525, 0.238 and 0.237 for acetic, propionic and butyric acids respectively on an early cut silage, while with a late cut silage the respective values were 0.548, 0.231 and 0.221 (Card and Schultz, 1953).

Luther and Trenkle (1967) fed lambs on a range of ground mixed diets containing from 1.0 to 0.2 parts of roughage and found that the molar proportions of acetic acid decreased from 0.695 to 0.439. Molar proportions of propionic acid in the rumen increased from 0.209 to 0.429 with decreasing roughage amount. Eusebio et al. (1959), Balch and Rowland (1957), Bath and Rook (1963), Beitz and Davis (1964) and Davis (1967) have reported decreased ruminal butyric acid with high concentrate diets. However, Luther and Trenkle (1967) found when a pelleted diet of 0.8 parts of concentrate was compared with one of 0.6 parts, the level of butyric acid in the rumen increased. These increases in butyric acid were also reported by Card and Schultz (1953), Elliot and Loosli (1959), Putnam et al. (1966) and Templeton and Dyer (1967) with high concentrate diets.

Several workers, Bath and Rook (1963), Topps and Elliot (1964), Coppock et al. (1964), Clanton and Woods (1966) and Templeton and Dyer (1967) have compared diets of 1.0 of roughage with others containing 0.5 of concentrate. Molar proportions of acetic acid in the rumen contents of animals fed the diets containing concentrate

were generally 0.06 units lower. The decline was compensated by increased proportions of propionic and butyric acids.

Coppock et al. (1964) supplied 1.0, 0.75 and 0.50 parts of the dietary energy as roughage, while Elliot and Loosli (1959) supplied 0.6, 0.4 and 0.2. The molar proportions of acetic acid in the rumen of cows sampled at four hours post-feeding in the former were 0.714, 0.682 and 0.653 respectively, while the latter workers reported 0.666, 0.647 and 0.605 respectively.

Ghorban et al. (1966) replaced hay by flaked corn and quoted mean values for the acetic acid proportion in rumen contents of 0.713 for the former and 0.420 for the latter diet, while corresponding values of propionic acid were 0.154 and 0.405 respectively.

El-Shazly (1952) fed diets of frozen fresh grass and dried grass to sheep and sampled rumen contents before and after feeding. He showed higher levels of acetate compensated by lower butyrate levels. The post-feeding fall in acetate concentration was 0.12 units for the fresh grass compared with 0.11 units for the dried grass. Ghorban et al. (1966) compared the effect of heat treatment on an all roughage diet. They quoted mean values for the molar proportions of acetic, propionic and butyric acids in the rumen of cattle of 0.712 and 0.657, 0.154 and 0.213 and 0.091 and 0.097 for diets of alfalfa hay and pelleted, dehydrated alfalfa respectively.

King and Hemken (1962) showed a decrease in the molar proportion of acetic acid in the rumen from 0.524 to 0.484 and a corresponding increase in the proportion of propionic acid from 0.276 to 0.322 with diets containing pelleted roughage together with either unheated or heated corn. Ensor et al. (1959) reported similar effects with heated concentrates. Clanton and Woods (1966) used diets of pelleted corn plus pelleted roughage, and cracked corn with pelleted roughage, and reported a decrease from 0.550 to 0.509 in the molar proportion of acetic acid in the rumen when the corn was pelleted. When pelleted hay was replaced by long hay heat

treatment of the corn produced no effect on the distribution of volatile fatty acids in the rumen (King and Hemken, 1962).

A mixed diet of silage, hay and flaked or cracked corn showed lower ruminal proportions of acetic acid and higher propionic and butyric acid levels for the flaked corn ration (Ghorban et al., 1966).

D. 4) Ammonia

Hogan and Weston (1967) reported mean values of 240 and 190 mg/l of ammonia nitrogen (NH_3-N) for the rumen contents of sheep given diets of chopped and ground lucerne hay and 11.4 and 3.4 mg/l for chopped and ground wheaten hay respectively. Oltjen et al. (1965) fed a diet containing 0.65 of hay to calves and found 164 and 152 mg/l of ammonia nitrogen in the rumen contents when the diet was unpelleted and pelleted respectively.

Annison et al. (1959) transferred a sheep receiving a diet of hay to lush spring grass and rumen ammonia nitrogen concentrations increased from a range of 59 to 211 mg/l to 484 to 562 mg/l. Johns (1955) observed rumen ammonia nitrogen concentration of sheep on a high protein pasture to range from 115 to 1300 mg/l, while Williams and Christian (1956) reported a range from 91 to 466 mg/l of ammonia nitrogen for grazing sheep. Later, the same authors (1966) studied the seasonal effect of a ryegrass plus white clover diet on the ammonia concentration in the rumen of sheep fed indoors at two levels of intake. At the low level of intake ammonia nitrogen concentrations (mg/l) were 185, 160, 324, 295 and 239 for December, March, April, June and September and, for the higher intake, 228, 208, 385, 340 and 310 respectively. Williams and Christian (1959) reported ruminal ammonia concentration of sheep at two hours post-feeding to range from 129 to 325 mg/l of ammonia nitrogen for the silage diets.

Briggs et al. (1957) examined the rumen ammonia levels of sheep on various diets, of roughage alone, roughages supplemented with carbohydrate concentrate and roughages supplemented with a carbohydrate and protein concentrate, and reported a range of values for rumen ammonia nitrogen concentrations of 20 to 2230 mg/l. When 0.5 of an all roughage diet was replaced by cracked maize, the concentration of ammonia nitrogen in the rumen at minimum values decreased from approximately 130

to 40 mg/l, while at maximum concentration the values were about 560 to 410 mg/l. Annison et al. (1959) also replaced part of the diet by cracked maize and reported rumen ammonia nitrogen concentrations at two hours post-feeding of 210 and 252 mg/l for the all hay and hay plus grain diets respectively. Bull et al. (1965) found ammonia nitrogen concentrations of 93 and 99 mg/l in the rumen contents of calves fed diets containing 0.5 and 0.2 of hay. When an all wheat diet was replaced by lucerne, the ammonia concentration in the rumen of sheep changed from 208 to 250 mg/l of ammonia nitrogen (Chou and Walker, 1964).

Chalmers (1963) gave the maximum concentration of ruminal ammonia nitrogen as approximately 400 and 340 mg/l for frozen and dried grass respectively. Christian and Williams (1957) quoted ruminal ammonia concentrations for sheep fed fresh and dried grass of 182 and 241 mg/l of ammonia nitrogen at pre-feeding and at post-feeding 292 and 221 mg/l respectively.

Chalmers (1963) has shown that processing of a dietary supplement can affect ammonia concentration in the rumen. When air dried groundnut meal was compared with toasted groundnut meal, maximum rumen ammonia nitrogen concentrations were 450 and 250 mg/l respectively. Davis and Stallcup (1967) with bullocks on diets of corn gluten feed, soyabean meal and raw soyabean reported rumen ammonia nitrogen concentrations before feeding of 140, 150 and 90 mg/l. Maximum concentrations occurred at one hour post-feeding for the corn gluten feed and three hours post-feeding for the other two diets. Maximum values were 240, 280 and 135 mg/l respectively. Sherrod and Tillman (1962) showed decreased maximum rumen ammonia concentration when diets of cotton seed meal were autoclaved compared with unheated cotton seed meal.

Annison et al. (1954) showed that maximum ruminal ammonia concentrations were reduced with diets containing groundnut or herring meal if the proportion of starch or cereal in the diet was increased.

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E. EFFECT OF TIME OF SAMPLING ON RUMEN PARAMETERS

1) Acids

The contents of the rumen are not of a constant composition and the pH, total volatile fatty acid (TVFA) concentration and the molar proportions of the individual fatty acids vary with their time of measurement relative to the time of ingestion of food. These variations have been investigated by several workers covering a range of diets.

Gray and Pilgrim (1951) fed alfalfa and wheaten hays, once daily, to sheep and sampled rumen contents at twelve intervals during twenty four hours. Minimum TVFA concentrations of 93 and 87 m mol/l were reported for the pre-feeding samples on the alfalfa and wheaten hay diets respectively and maximum concentration for the alfalfa of 255 m mol/l at four hours and, for the wheaten hay, 205 m mol/l at six hours post-feeding. The distribution of acetic, propionic and butyric acids in the rumen with the alfalfa diet at the pre-feeding sampling was 0.70, 0.15 and 0.15 respectively and, at four hours post-feeding 0.69, 0.19 and 0.12 respectively. The proportion of acetic acid was highest at 0.73 at twelve and fourteen hours post-feeding. With the wheaten hay diet the distribution of acetic, propionic and butyric acids in the rumen at pre-feeding was 0.68, 0.18 and 0.14 respectively, and 0.58, 0.26 and 0.16 respectively four hours later, when the proportion of acetic acid was minimal.

Ensor (1959, cited by Shaw, 1961) with cows on an alfalfa hay diet quoted rumen TVFA concentrations of 8.47, 8.51, 7.54 and 6.38 g/l when the rumen contents were sampled at two, five, eight and eleven hours after feeding. The range of molar proportions of the rumen acids was 0.693 to 0.724 for acetic, 0.164 to 0.181 for propionic and 0.079 to 0.105 for butyric. Annison et al. (1959) have also shown variation in rumen characteristics with time of sampling after feeding in animals

fed hay alone.

Ghorban et al. (1966), with an alfalfa hay diet, sampled rumen contents at hourly intervals from pre-feeding to six hours after feeding and found the concentration of total volatile fatty acids in the rumen varied with the time of sampling, and ranged from 80 to 105 m mol/l.

Fenner et al. (1967) fed hay twice daily to cows and sampled the rumen contents at hourly intervals for nine hours and reported statistically significant differences between hourly samples for the rumen parameters of pH, TVFA concentration and the molar proportions of acetic, propionic and butyric acids.

For diets of fresh grass Christian and Williams (1957) reported a maximum rumen pH value of 7.2 at pre-feeding and a minimum value of 6.7 at three hours post-feeding. Terry and Tilley (1964) reported a maximum pre-feeding value of 6.9 and a minimum of 5.9 at two, four and six hours post-feeding. At pre-feeding the molar proportions of acetic, propionic and butyric acids were 0.71, 0.16 and 0.12 respectively, and, when that of acetic acid was minimal at four hours post-feeding the distribution was 0.53, 0.28 and 0.19 respectively.

Williams and Christian (1959) fed four silages to sheep twice daily and sampled rumen contents at one and two hours pre-feeding and at six one hourly intervals thereafter. At pre-feeding the concentrations of TVFA were between 15 and 40 m mol/l and these increased rapidly to maximum values between 49 and 63 m mol/l at one hour post-feeding. They then fell sharply.

When a conventional roughage plus concentrate diet was fed at twelve hour intervals, and the rumen contents sampled at twelve intervals, Schambye and Phillipson (1949) reported maximum rumen pH of 6.7 at pre-feeding and at nine and ten hours after feeding, and at a minimum of 6.2 two to three hours post-feeding. The acid concentration was at a minimum of 70 m mol/l at pre-feeding and at a maximum of 127

m mol/l two hours later. The distribution of acetic, propionic and butyric acids at pre-feeding was 0.742, 0.157 and 0.101 respectively, while at one hour post-feeding was 0.706, 0.189 and 0.105 respectively. Several workers, notably Balch and Rowland (1957), Moir and Somers (1957) and Simkins et al. (1965), have confirmed a similar pattern of rumen changes for mixed roughage and concentrate diets. Clanton and Woods (1966) with steers fed pelleted diets, differing in the amount of concentrate inclusion, could find no pattern of change with the time of sampling rumen contents relative to feeding. Putnam et al. (1966), with pelleted and ground diets, reported that the concentration of TVFA was maximal at one hour post-feeding for both diets, and values ranged from 70 to 89 m mol/l for the ground diet and from 74 to 106 m mol/l for the pellets.

Balch and Rowland (1957) sampled hourly the rumen contents of cows fed high concentrate diets twice daily. Maximum level of pH was at pre-feeding and the minimum value at two to four hours post-feeding. The range of values was from 4.79 to 6.89. The minimum concentration of total volatile fatty acids was at pre-feeding and the maximum at four hours post-feeding with a range of concentration from 67 to 166 m mol/l. The range of molar proportions of acetic, propionic and butyric acids in the rumen was from 0.398 to 0.413, 0.369 to 0.378 and from 0.081 to 0.106 respectively.

Briggs et al. (1957) with a wide variety of diets, some supplemented with protein concentrate as well as a carbohydrate concentrate, fed sheep once daily and sampled the rumen contents at frequent intervals between feeds. Minimum values for rumen TVFA concentration and maximum values for pH were obtained with pre-feeding samples for all diets, but the time of sampling for maximum acid concentration and minimum pH value varied with the diet.

E. 2) Ammonia

Briggs et al. (1957) fed a diet of lucerne chaff to sheep and reported minimum values for rumen ammonia nitrogen concentration of 180 mg/l at pre-feeding and maximum concentrations of 400 to 450 mg/l between two and three hours post-feeding. Annison et al. (1959) with an all hay diet showed a similar pattern.

Rumen ammonia nitrogen concentration in sheep fed diets of fresh grass showed minimum pre-feeding values of 180 mg/l (Christian and Williams, 1957) and 280 mg/l (El Shazly, 1952), with corresponding maximum values at one to two hours post-feeding of 290 and 390 mg/l. When dried grass was given El Shazly (1952) reported a similar rumen ammonia concentration pattern with time of sampling with values ranging from 210 to 300 mg/l. Christian and Williams (1957) reported pre-feeding values of 240 mg/l and maximum post-feeding values of 246 mg/l at one hour after feeding.

Williams and Christian (1959) with four silages of differing ammonia nitrogen contents showed low concentration of rumen ammonia nitrogen at pre-feeding followed by a sharp rise to maximum concentration at one hour post-feeding. The concentration then fell to minimum levels at six hours post-feeding. The pattern of change was similar for all the silages although the maximum concentration varied between 170 and 250 mg/l.

Moir and Somers (1957) and Somers (1961) fed mixed concentrate roughage diets, once daily, to sheep and sampled the rumen contents at two-hourly intervals for twenty-four hours. Minimum concentration of ruminal ammonia occurred from sixteen hours post-feeding and rose sharply after feeding to maximum concentration at four hours post-feeding.

Annison et al. (1954) found similar patterns of rumen ammonia concentration when rumen contents were sampled at twelve one-hour intervals, after feeding a

basic diet supplemented with casein or groundnut. However, with a flaked maize supplement a different rumen concentration curve was obtained. No definite maximum was shown; pre-feeding concentrations, for two sheep, were about 100 and 180 mg/l which fell to between 10 and 20 mg/l two to three hours afterward. This type of curve was also obtained when the hay was supplemented with ground or flaked maize. Briggs et al. (1957) with a diet of lucerne and cracked maize reported changes in rumen ammonia concentration to be minimal at pre-feeding and maximal at four hours post-feeding.

Sherrod and Tillman (1962) sampled rumen contents at pre-feeding and at hourly intervals for twelve hours when sheep were given diets containing processed soyabean meal and soyabean meal heated for forty-five and ninety minutes. The concentration of ammonia in the rumen varied with the time of sampling for all the diets. Minimum concentrations were in the pre-feeding samples and maximum occurred between two and three hours after feeding.

F. RUMEN FERMENTATION AND NUTRITIVE VALUE.

1) Utilization of Rumen Fermentation Products for Maintenance

Armstrong and Blaxter (1957a) from calorimetry trials reported efficiencies of utilization for maintenance of 0.592, 0.865 and 0.764 for acetic, propionic and butyric acids respectively when the acids were continuously infused into the rumen of sheep to supply 2.93 MJ/day. The efficiency was 0.875 when a three component mixture of the volatile fatty acids was used. Armstrong et al. (1957) found the efficiency was increased to 0.907 with the infusion of a mixture of propionic and butyric acids in the ratio of 3 to 2. When an acid mixture of acetic 0.25, propionic 0.45 and butyric 0.30 was used the efficiency decreased to 0.872 and fell to 0.847 when the acetic acid was increased to 0.9 of the mixture. Blaxter (1962) gave a value of 0.85 for the efficiency of utilization for maintenance of a mixture of the three major volatile fatty acids similar to that found in rumen contents.

Armstrong (1964) quoted efficiencies of utilization of the metabolisable energy of dried grass for maintenance of 0.78 when the molar proportions of acetic, propionic and butyric acids were 0.63, 0.23 and 0.15, and of 0.67 when the proportions were 0.70, 0.19 and 0.10. An efficiency of 0.72 was reported when the molar proportions of the acids were 0.715, 0.180 and 0.105 and 0.695, 0.205 and 0.100. With each species of grass the first cut had the highest efficiency of utilization of energy for maintenance and this declined for each subsequent cut, while the molar proportion of acetic acid in the rumen was lowest for the first cut and increased with each subsequent cut.

Blaxter and Wainman (1964) reported efficiencies of utilization of metabolisable energy for maintenance of 0.71 and 0.70 for sheep and cattle respectively, when the molar proportions of acetic and propionic acid in the rumen were 0.70 and 0.20. When the molar proportions were 0.50 and 0.23 the efficiency values were

0.81 and 0.75.

Corbett et al. (1966) fed sheep early and late cut grass at eight and nine levels of gross energy intake. To avoid undue bias by an arbitrary choice of mathematical model to relate metabolisable energy intake with energy retention they fitted four sets of equations to their data to derive estimates of the requirement of metabolisable energy for maintenance and quoted 0.68 and 0.60, 0.70 and 0.63, 0.70 and 0.70 and 0.73 and 0.67 for the early and late cuts when the molar proportions of acetic, propionic and butyric acids in the rumen were 0.66, 0.24 and 0.07 respectively and 0.72, 0.17 and 0.08 respectively.

F. 2) Utilization of Rumen Fermentation Products for Growth and Fattening

Armstrong and Blaxter (1957b) gave supplements of the individual volatile fatty acids by continuous intraruminal infusion to sheep on maintenance rations of dried grass. There were no statistically significant changes in rumen acid concentrations. The efficiencies of utilization for lipogenesis of acetic, propionic and butyric acids were 0.33, 0.56 and 0.62 respectively. When mixtures of acetic, propionic and butyric acids, in the ratio of 75 to 15 to 10 and 25 to 45 to 30, were infused with the same basic maintenance diet of dried grass, the efficiency of utilization of the energy of the mixtures for lipogenesis was 0.32 and 0.58 respectively (Armstrong et al., 1958).

The efficiency of utilization of the metabolisable energy of S23 ryegrass declined from 0.525 to 0.326 when the molar proportions of acetic acid in the rumen increased from 0.627 to 0.686. Similar patterns were obtained with other grass varieties (Armstrong, 1964).

Corbett et al. (1966), working with sheep fed early and late cut grass, reported molar proportions of acetic, propionic and butyric acids in the rumen of 0.66, 0.24 and 0.07, and 0.72, 0.17 and 0.08 respectively. Efficiencies of utilization of energy for fattening of 0.45 and 0.42, 0.46 and 0.34, 0.44 and 0.28, and 0.41 and 0.31 were quoted since, as was the case with the efficiency of utilization of these diets for maintenance; there was not sufficiently extensive data to justify these authors concluding which of the four mathematical models relating energy retention with metabolisable energy intake was closest to the biological facts.

Blaxter and Wainman (1964), with sheep and steers, found the efficiency of utilization of metabolisable energy for lipogenesis increased from 0.32 to 0.62 and from 0.28 to 0.59 respectively, when the molar proportions of acetic, propionic and butyric acids in the rumen were changed from 0.70, 0.20, 0.10 to 0.45, 0.42 and 0.04.

Rook et al. (1963) infused acetic, propionic and butyric acids into the rumen of heifers and reported increased rumen volatile fatty acid concentration, but very little effect on the ratios of volatile fatty acids other than the one infused. They also showed increased empty body weight gains, which were only significant at the higher rates of infusion. Butyric acid was the most effective but acetic and propionic acids were not significantly different.

Addition of salts of the volatile fatty acids to lamb rations did not significantly alter the rates of body weight gain (Essig et al., 1959; Nicolson et al., 1964), nor did the addition of sodium propionate to beef cattle rations (Nicolson et al., 1961). Elliot et al. (1965) reported that the addition of sodium salts of acetic or propionic acids to a basic hay ration greatly increased the efficiency with which the energy was stored in the growing-fattening lamb; but there was no difference between the supplements.

Orskov and Allen (1966a) could show no significant differences in the promotion of live weight gain, empty body weight or carcass weight, when ruminal infusions of salts of the volatile fatty acids were used to achieve varying molar proportions of the major volatile fatty acids in the rumen. The discrepancy found in the efficiency of utilization of the energy of the acids for fattening by carcass analysis and by calorimetry was investigated by Orskov et al. (1966) and Orskov and Allen (1966b,c). Neither the stage of maturity of the experimental animals, nor the basic diet, nor the frequency of feeding were responsible for the differences.

The "ideal" ratio of acetic to propionic acid in the rumen of steers has been investigated by many workers. Shaw et al. (1960) reported an increase in daily weight gain from 0.9 to 1.1 kg. when the ratio of acetic to propionic acid was reduced from 4.2 to 1.1. Weiss et al. (1967) reported increased daily weight gains of 0.89, 0.85 and 1.1 kg. when the ratios were 3.4, 2.8 and 1.3 respectively. Newland et al. (1962), Ekern and Reid (1963) and Oltjen et al. (1966) reported

similar findings. Putnam et al. (1965) attempted to relate rumen TVFA concentration with daily weight gain but the relationship could only explain 0.11 to 0.14 of the total variation.

Grimes et al. (1967) found that 0.48 of the variation in live weight gain of grazing lambs could be accounted for by differences in the molar proportions of acetic and propionic acids in the rumen.

F. 3) Utilization of Rumen Fermentation Products for Milk Production

Rook and Balch (1961) and Rook et al. (1965) measured milk production in dairy cows receiving a diet of hay plus concentrates when acetic, propionic and butyric acids were infused intraruminally. Acetic acid increased the yield of milk, of milk fat and the milk fat content. Propionic acid decreased and butyric acid increased the yield of fat and the content of fat, but neither acid affected milk yield. With a combination of acids infused there was no effect on milk yield. Propionic acid alone or with acetic acid again reduced the milk fat content. Wilson et al. (1967), in a similar intraruminal infusion experiment, reported increased yield of milk with each acid infusion and increased yield of fat with both acetic and butyric acid infusions. The former did not affect the composition of the milk but the latter increased the fat content.

Feeding sodium propionate (Schmidt and Schultz, 1958) or sodium acetate (Balch et al., 1961) produced no significant change in yield or fat content of milk, although the total acid content of the rumen was increased. When low molar proportions of acetic and high proportions of propionic acid were produced in the rumen the addition of sodium acetate to the diet improved the fat content of the milk (Balch and Rowland, 1959; Stanley et al., 1964).

Balch et al. (1967) added calcium salts of acetic, propionic and butyric acids to the diets of cows and in all cases found slightly increased milk yields but decreased fat content and concluded that although relative effects were similar, the different results, from dietary addition of salts and acid infusions, were due to additional calcium in the diet. Rook and Line (1961) suggested increased yield of milk resulted from increased production of acetic and propionic acids in the rumen and gave evidence to show that the acids differed in their effect on the synthesis of milk constituents, acetic affecting both lactose and protein, while propionic

affected protein.

A decrease in the fat content of the milk of cows at the same stage of lactation is usually accompanied by a decrease in the rumen in the proportion of acetic acid and an increase in propionic acid (Tyznik and Allen, 1951; Balch et al., 1955; Shaw, 1961; Hawkins et al., 1963; Beitz and Davis, 1964; Storry and Rook, 1965; Huber and Boman, 1966 and Hawkins and Little, 1967). Davis (1967) measured acetic acid production and reported 29.3 and 28.1 mol/24h when the milk fat content was 15.6 and 32.3 g/kg respectively and concluded that an absolute deficit of acetic acid production was not responsible for the decline in milk fat. Ensor et al. (1959) and Jorgensen and Schultz (1963) found a decrease in the molar proportions of acetic and an increase in propionic acids in the rumen accompanied a decrease in milk production, as well as a decrease in milk fat content. Stanley et al. (1964) and King and Hemken (1962) reported that milk production did not change with a decrease in the proportion of acetic to propionic acids in the rumen contents, but confirmed the decline in milk fat content. Colenbrander et al. (1967) reported increased milk production but decreased production of fat corrected milk when the ratio of acetic to propionic acids in the rumen declined from 4.0 to 1.8. Yamdagni et al. (1967) and Hinders and Owen (1963) found very little difference in milk fat yield or content with a change in the ratio of rumen acids, but the latter workers quoted a change from 2.2 to 2.7.

Elliot and Loosli (1959) reported that the efficiency with which digestible energy was converted to fat corrected milk was highly correlated with the relative proportions of propionic acid in the rumen volatile fatty acids and with the ratio of acetic to propionic acids. Blaxter (1962), using data from Elliot and Loosli (1959), reported that molar proportions of acetic acid between 0.55 and 0.62 were optimal for maximum efficiency of utilization of metabolisable energy for milk production above maintenance.

Coppock et al. (1964) related lactation efficiency to the proportions of volatile fatty acids in the rumen, r = -0.73 for acetic acid and +0.43 for propionic acid. Storry and Rook (1966) found 0.60 of the variation in milk fat content was associated with increased ruminal propionic acid.

Armstrong and Blaxter (1965) infused acetic and propionic acids and a mixture of acetic 0.55, propionic 0.32 and butyric acids 0.13 continuously into the rumen of lactating goats on a hay plus concentrate diet. The efficiencies of utilization of metabolisable energy for lactation at zero energy balance were 0.650, 0.723 and 0.714 respectively.

III

METHODS

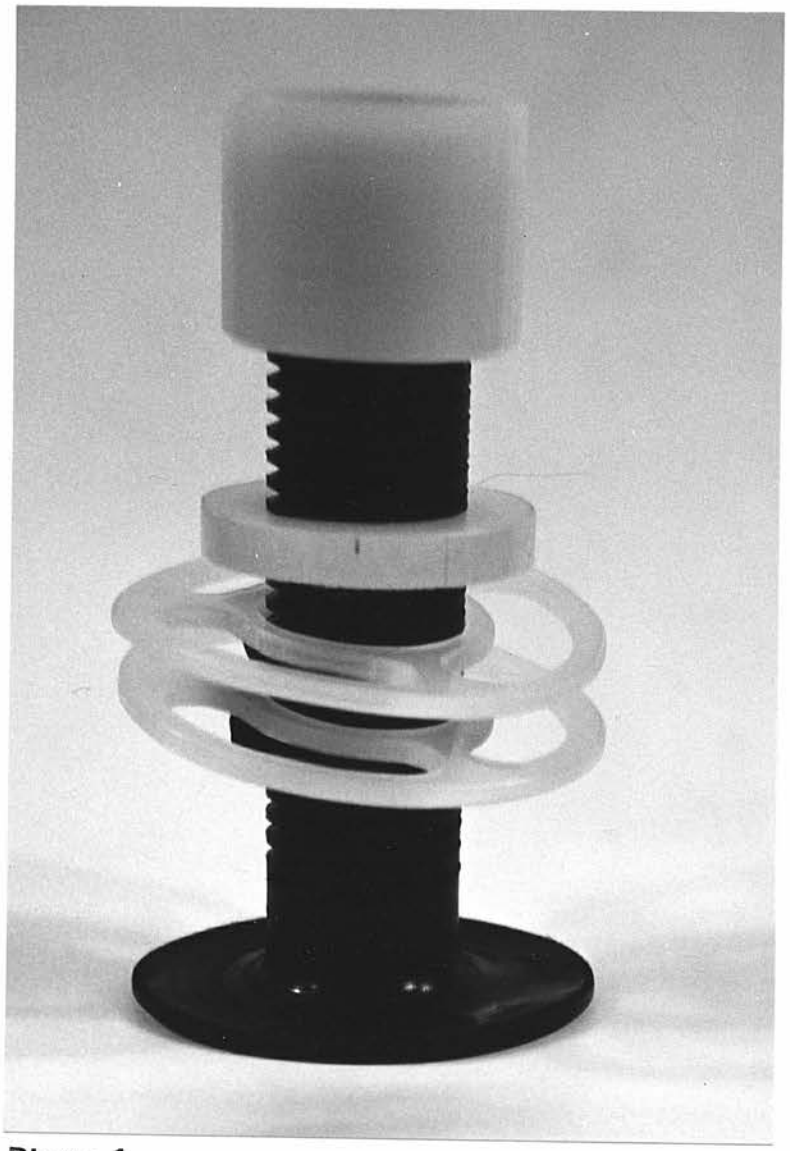

Plate 1

A. EXPERIMENTAL.

1) Animals

Eighteen month old, half bred, Cheviot wethers were fitted with permanent rumen cannulae similar to the type used by Alexander (1970) and illustrated in Plate 1. In all the trials each sheep received each treatment.

A. 2) Trial Procedure

In general the techniques employed were the same for each experiment. The exceptions are detailed in the text.

Seven to fourteen days before the commencement of a trial the sheep were introduced to silage, preferably one of different composition to the experimental materials. When the introduction to silage was complete, the sheep were weighed, harnessed for collection of faeces and urine and placed in metabolism crates (McDonald, 1958). After two to three days, to allow the animal to adapt to the crate, the feeding regime was started. The ration was given in equal feeds at 09.00 and 21.00 h. daily, and residues were removed at 11.00 and 23.00 h. respectively. The experimental diets were introduced gradually, and when intakes had stabilised, usually in about seven days, the eight day digestibility and metabolism trials were initiated. Apparent digestibilities of dry matter, organic matter and nitrogen of the foods were measured. Metabolisable energy values were calculated from digestible energy using measured urinary energy loss and methane loss calculated according to the equation of Blaxter and Clapperton (1965). The sheep were weighed, prior to feeding, at the beginning and end of each digestibility period. Dry matter intakes were recorded at each feed and separate oven dry matter determinations were carried out on the residues from each sheep. The true dry matter intake of silage was calculated by applying a factor, based on dry matter determined by

toluene distillation, to the dry matter of the residue. Urine was collected in sulphuric acid (25 ml containing 6.25 ml concentrated acid, daily). The total volume was measured at the end of each period and an aliquot (0.025) retained for the determination of nitrogen and gross energy. Faeces were collected daily over the eight day period with a three day time lag between commencement of intake measurements and the first collection of faeces (Raymond et al., 1953). Faeces were stored at -20°C until the end of the trial period. After allowing the samples to defreeze, the total weight was recorded, and a sub-sample (0.1) obtained by "quartering". This was retained for determination of dry matter, nitrogen, ash and gross energy.

After a changeover period of three to four days, during which the new diet gradually replaced the old, the second phase of the experiment started. A third and perhaps a fourth period followed, depending on the number of foods under investigation.

During the trials water was available to the sheep at all times and fresh water was offered daily with each feed. A mineral plus vitamin supplement was given with each food in a quantity sufficient to meet the maintenance requirement of the sheep (Agricultural Research Council, 1965).

A. 3) Sampling

a. Rumen: Upon completion of each eight day digestibility period the rumen contents were sampled at 08.50, 10.00, 11.00, 12.00, 13.00, 15.00, 17.00, 19.00 and 21.00 h. on two consecutive days. In no experiment were the rumen contents sampled before the sheep had been given the diet for at least fourteen days (Whittenbury, 1969). At each sampling 100 ml of rumen contents were withdrawn from each sheep, through semi-rigid polythene tubing (1 m x 8 mm id) using a suction pump. The end

of the tube inserted into the rumen was perforated for about 40 mm with holes 4 mm in diameter and was cut obliquely. The sample was taken from several regions of the rumen by repeated insertion and withdrawal movements of the tube. PH values were determined immediately. The sample was filtered through four layers of butter muslin and a 5 ml aliquot of the liquor taken for measurement of ruminal ammonia concentration. 2 ml of saturated mercuric chloride solution was added to the remainder of the liquor (Edwards, 1964), which was then stored at -20^{o} to await subsequent analysis.

A. 3) b. Blood: On the day following the sampling of rumen contents blood was taken, by jugular puncture, three hours after feeding, when differences in blood pH, glucose and urea might be expected to be most obvious (Lewis, 1957; Rook and Line, 1961; Hawkins et al., 1970; Ross and Kitts, 1973; Thye et al., 1970). The sample for the determination of pH was taken anaerobically using a 5 or 10 ml syringe with a Luer fitting. The needle was inserted into the jugular vein and, when the blood was flowing freely, the syringe, containing heparin in the neck, was attached and the blood allowed to fill the barrel. When filled, the syringe was removed from the needle, a small quantity of blood was ejected from the end of the syringe which was then sealed immediately with a cap containing heparin, then thoroughly mixed (Littlejohn, 1969). The pH was determined immediately, or within forty-five minutes if kept in an ice bath. A further sample was taken using heparin as anti-coagulant (Annison, 1954b), and this was immediately centrifuged at 3000 rpm for fifteen minutes (Hawk and Bergeim, 1931). The plasma was stored at -20^{o} for the subsequent determination of glucose and urea.

A. 4) Foods

The total amount of any one silage necessary for a complete trial was taken

at one time from one source, in most cases a two-tonne capacity polyplastron silo.[1] After discarding any waste material, the silage was well mixed and a weighed amount, sufficient for one meal, was placed, with an identification label, in a heavy gauge (500) polythene bag. The silage was manually compressed, to exclude air, sealed with rubber bands and identification tags attached. The feeds were stored at -20° until twenty-four to thirty-six hours before feeding. During the filling of the bags a bulk sample, for chemical analysis, was taken by random grab sampling of the mass.

When frozen fresh grass was used as a treatment it was cut with a double chop forage harvester from the same field, on the same day, at approximately the same time as the grass used to make the silage. It was sampled, bagged and stored as for the silage.

When the grass was dried and used as a treatment the conditions of cutting were as for frozen fresh grass. The grass was dried with air at 45° at a constant velocity until the required dry matter of about 900 g/kg was achieved. The drying equipment described by Clark (1966) was used. The grass in the bins was frequently turned during the drying process.

The dried grass was mixed and sampled as for silage and a quantity sufficient for one meal was weighed into a polythene bag. After sealing, with rubber bands, the grass was stored at room temperature until fed. The complete pelleted diet was sampled, bagged and stored under similar conditions.

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1. Manufactured by Gordon Low, Isle of Wight.

B. ANALYTICAL.

1) Blood

a. pH: Blood pH values were recorded on a Pye 78 pH meter using a micro-electrode unit (135N / 335N / 945). Blood was aspirated directly from the syringe into the electrode chamber maintained at 38° by a water circulation thermostat and when the system was completely flushed out the pH was read.

B. 1) b. Plasma Glucose: Glucose was determined by auto-analyser, using the macro method of Trinder (1969). In this method glucose is oxidised in the presence of glucose oxidase to hydrogen peroxide. This is then allowed to react, in the presence of peroxidase, with the colourless oxygen acceptor 4-aminophenazone. The colour developed is proportional to the concentration of glucose in the original solution.

B. 1) c. Plasma Urea: Urea was determined using the Technicon Autoanalyser Method No-1c, based on the procedure of Marsh et al. (1965). Urea reacts with diacetyl monoxide in the presence of thiosemicarbazide in acid conditions to give a red colour which is directly related to the concentration of urea.

B. 2) Rumen

a. pH: PH values were determined using a Pye 78 pH meter with a Pye Ingold combined electrode.

B. 2) b. Volatile Fatty Acids: 10 ml of strained rumen contents, at 0° to 4°C, were mixed with 2 ml metaphosphoric acid (250 g/l) in sulphuric acid (5N) (Packett and McCune, 1965). After thirty minutes refrigeration the mixture was centrifuged

at 3500 rpm for thirty minutes, refrigerated for a further thirty minutes and the clear, deproteinised, supernatent liquid either stored at -20^{o} to await analysis or used immediately.

The volatile fatty acids in rumen contents were estimated using gas liquid chromatography (GLC). Earlier determinations were carried out using an ether extraction procedure described by Edwards (1967). Later determinations used direct injections of the acidified, deproteinised rumen liquor on to the column. Two gas chromatographs and three columns were used for the determinations.

Table 8

Details of GLC Separations of Ruminal Volatile Fatty Acids

Instrument	Perkin-Elmer F11	Pye 104	Perkin-Elmer F11
Nature of sample	in ether	in water	in water
Size of sample	2 µl	0.7 µl	2 µl
Detector	Flame ionization	Flame ionization	Flame ionization
Column material	stainless steel	glass	glass
Column size	2m x 3mm od	1.5m x 4mm od	1.8m x 6mm od
Stationary phase	FFAP(1)	neopentyl glycol adipate	FFAP(1)
Support	Chromosorb G. (80–100 mesh)	Diatomite C (100–120 mesh)	Chromosorb G. (80–100 mesh)
Amount of stationary phase	0.05	0.20	0.05
Carrier gas	Nitrogen	Argon	Argon
Carrier gas pressure (bar)	0.90	1.21	1.52
Hydrogen gas pressure (bar)	1.03	0.69	0.97
Air pressure (bar)	1.72	1.72	1.72

Table 8 (Cont'd)

Details of GLC Separations of Ruminal Volatile Fatty Acids

Column temperature (°C)	140	127	145
Injection block temperature (°C)	175	0	175
Recorder	Kent Mark 3 (1 sec. response)	Leeds & Northrup Speedomax W (1.2 sec. response)	Kent Mark 3 (1 sec. response)
Chart speed	380 mm/h	254 mm/h	380 mm/h
Attenuation	C_2-C_4 (2) $2x10^2$	C_2 5×10^2	C_2 5×10^2
	C_5+C_6 20x1	C_3+C_4 2×10^2	C_3+C_4 2×10^2
		C_5+C_6 50×1	C_5+C_6 50×1

Response was linear over the concentration range of each acid. Repeatability with each instrument and reproducibility between the columns and instruments was within 0.01 for the major acids. No deterioration of samples occurred on storage at -20°, and the determined molar proportions of the acids did not change over several months storage.

A solution containing known amounts of the volatile fatty acids under investigation, subjected to the same deproteinisation and centrifugation techniques as the samples, was used as a standard. One standard was injected for every ten rumen liquor samples.

The higher minor acids of the rumen were present in small amounts, and to

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(1) Manufactured by Perkin-Elmer.
(2) C_2 acetic acid
C_3 propionic acid
C_4 butyric acids
C_5 valeric acids
C_6 hexoic acid.

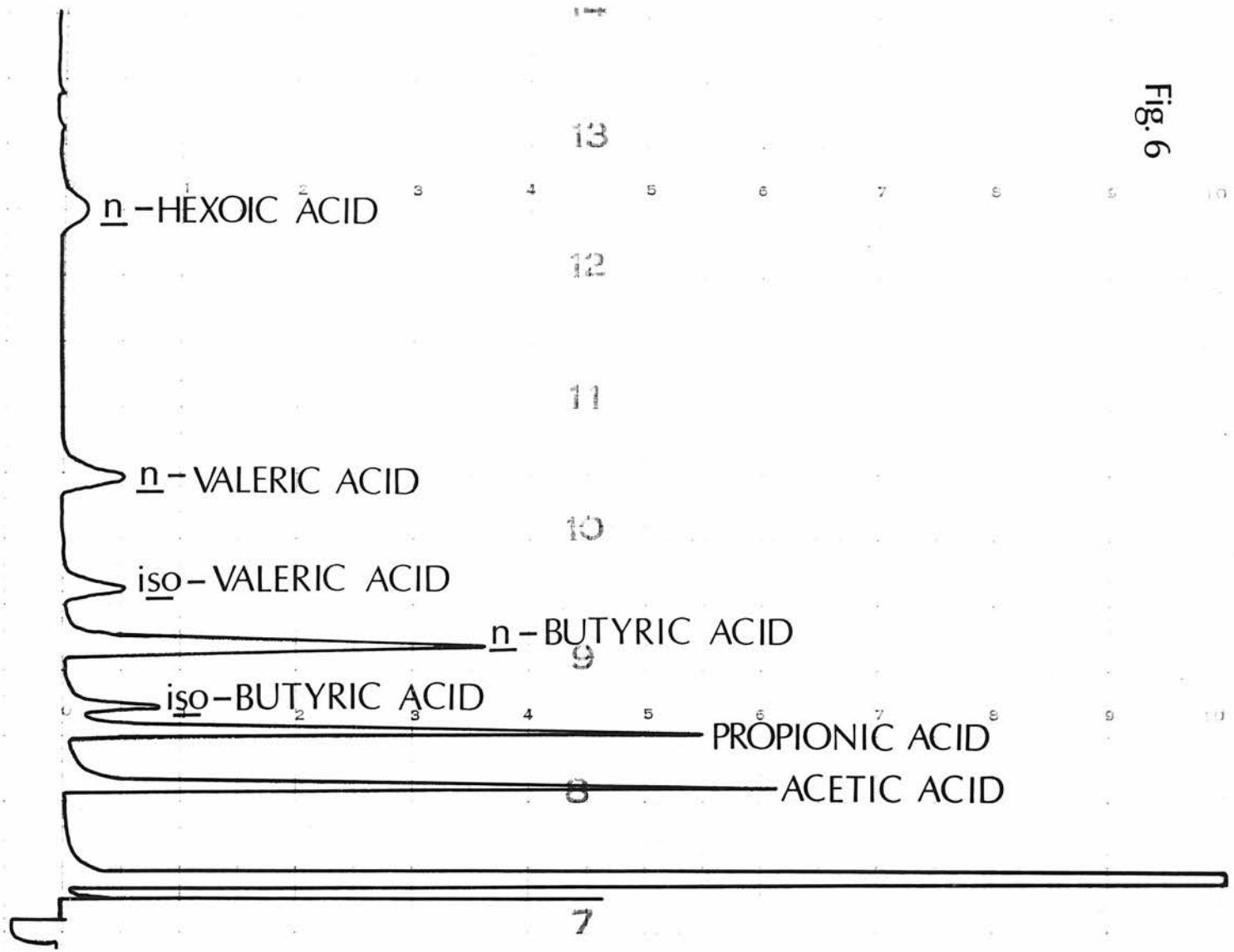
n-HEXOIC ACID
n-VALERIC ACID
iso-VALERIC ACID
n-BUTYRIC ACID
iso-BUTYRIC ACID
PROPIONIC ACID
ACETIC ACID

Fig. 6

increase the accuracy of their measurement the attenuation was increased. This resulted in a baseline unsuitable for integration measurements. In addition, the baseline did not always return to zero between the peaks for propionic and iso-butyric acids. Since the peaks were narrow and Gaussian, peak heights were measured and the concentration of the acids calculated by comparison with peak heights obtained with the standard solutions.

Fig. 6 illustrates a typical chromatogram obtained in the separation of the volatile fatty acids in a sample of rumen liquor.

B. 2) c. Ammonia: Ruminal ammonia concentration was determined in 5 ml of sample of strained rumen contents, using the microdiffusion technique of Conway and O'Malley as described by Chalmers et al. (1954) with modifications suggested by Chalmers (1968).

B. 2) d. Lactic Acid: Strained rumen liquor was mixed with zinc sulphate and barium hydroxide solutions to give a final dilution rate of 1 to 3, and filtered. Lactic acid was determined on 10 ml portions of protein free filtrate by the Nanni and Baldini (1965) modifications of the Barker and Summerson (1941) method. Lactic acid was oxidised, by sulphuric acid, to acetaldehyde, which was then determined colorimetrically by reaction with p-hydroxydiphenyl in the presence of copper.

B. 3) Foods

Samples of dried foods, hammer milled to pass through a 1mm screen, were used for analyses unless otherwise stated. The values for silages were then adjusted to a true dry matter basis using a correction factor based on the relationship between dry matter content determined by oven drying and by toluene distillation.

B. 3) a. Dry Matter: Dry matter was determinated by drying in a forced draught oven at 100°C for sixteen hours, or by toluene distillation using the method of Dewar and McDonald (1961).

B. 3) b. Modified Acid Detergent Fibre: Modified acid detergent (MAD) fibre was determined in 1g samples by the Clancy and Wilson (1966) modification of the Van Soest (1963) method.

B. 3) c. Cellulose: Cellulose was determined in 1g samples by the method of Crampton and Maynard (1938).

B. 3) d. Crude Fibre: Crude fibre was determined according to the Fertiliser and Feeding Stuffs Regulations (1968).

B. 3) e. Acids: The volatile fatty acid content of silage was determined after extraction of 25g of fresh silage with 0.6N sulphuric acid by the same gas chromatographic method used for ruminal fatty acids. Succinic acid, lactic acid and volatile fatty acids were measured in the silages by the silicic acid column chromatographic method of Lessard and McDonald (1966).

B. 3) f. Crude Protein: Total nitrogen was determined on the fresh silage and on dried milled samples of other foods by the Kjeldahl method using selenium / potassium sulphate catalyst. Crude protein was calculated by multiplying total nitrogen by 6.25.

B. 3) g. Total Soluble Nitrogen: Fresh silage (100g), fresh grass or dried grass was extracted with boiling water to a final volume of 1.5l and the nitrogen content

of an aliquot determined by a micro distillation procedure (Macpherson, 1968).

B. 3) h. Volatile Nitrogen: Volatile nitrogen content of silage was determined in an aliquot of the aqueous total soluble nitrogen extract. Sodium borate was added to bring the pH to about 9.2 and the ammonia nitrogen estimated by a micro distillation procedure.

B. 3) i. Water Soluble Carbohydrate: Water soluble carbohydrate content was determined by the method of McDonald and Henderson (1964).

B. 3) j. pH: The pH of fresh silage and grass was measured using an aqueous macerate of the food (25g of fresh silage in 200 ml water) using a Pye 78 pH meter as for rumen contents.

B. 3) k. Ash: Ash was determined according to the Fertiliser and Feeding Stuffs Regulations (1968) by ignition in a muffle furnace at 500°C.

B. 3) l. Buffering Capacity: The buffering capacity of the foods was determined according to the method of Playne and McDonald (1966).

B. 3) m. Ethanol: Ethanol was determined on fresh silage by the Kent-Jones and Taylor (1954) modification of the Kozelka and Hine (1941) method.

B. 3) n. Gross Energy: Gross energy was determined on fresh silage to prevent loss of energy by drying at 100° (Colovos et al., 1957) in an adiabatic bomb calorimeter using polythene as primer (McDonald et al., 1973).

B. 4) Faeces and Urine

a. Dry Matter: Faeces dry matter was determined by the method described for foods.

B. 4) b. Nitrogen: Nitrogen was determined by the method used for silage, with fresh material to prevent loss of volatile nitrogen by drying (Colovos et al., 1957; Siriwardene et al., 1966).

B. 4) c. Ash: The determination was as described under foods.

B. 4) d. Gross Energy: Gross energy was determined on fresh faeces samples as for silage. Urine samples were dried on polythene in a vacuum dessicator over sulphuric acid (Alderman et al., 1971).

IV

EXPERIMENT 1.

THE INVESTIGATION OF RUMEN FERMENTATION PATTERNS ON A SILAGE DIET

EXPERIMENTAL

Each of six fistulated sheep were given each of two diets in a simple cross-over design. The composition of the diets is given in Table 9. The pH of the silage was 4.09 and the dry matter 309 g/kg compared with 868 g/kg for the complete diet.

Table 9

Composition of the Silage and the Complete Diet (g/kg dry matter)

	Silage	Complete Diet
Total nitrogen	21.9	24.7
Crude fibre	282	172
MAD - fibre	320	-
Ether extract	27	35.7
Ash	103	97
Water soluble carbohydrate	116	-
Lactic acid	62	-
Succinic acid	8	-
Acetic acid	14	-
Propionic acid	0.8	-
Butyric acid	1.3	-

The wilted silage was made under farm conditions in June 1969 from a first cut of mixed ryegrasses. The complete diet was a commercial pellet of straw (0.30), cut into 15 mm lengths, combined with barley (0.22 to 0.25), molasses (0.10) and urea (0.01). The same weight of dry matter (1.32 kg) for both diets was offered daily in two equal feeds at twelve hour intervals. Access to each meal was restricted to two hours.

After the period of adaptation feed intakes were measured over a period of fourteen days. During the last forty-eight hours the contents of the rumen were

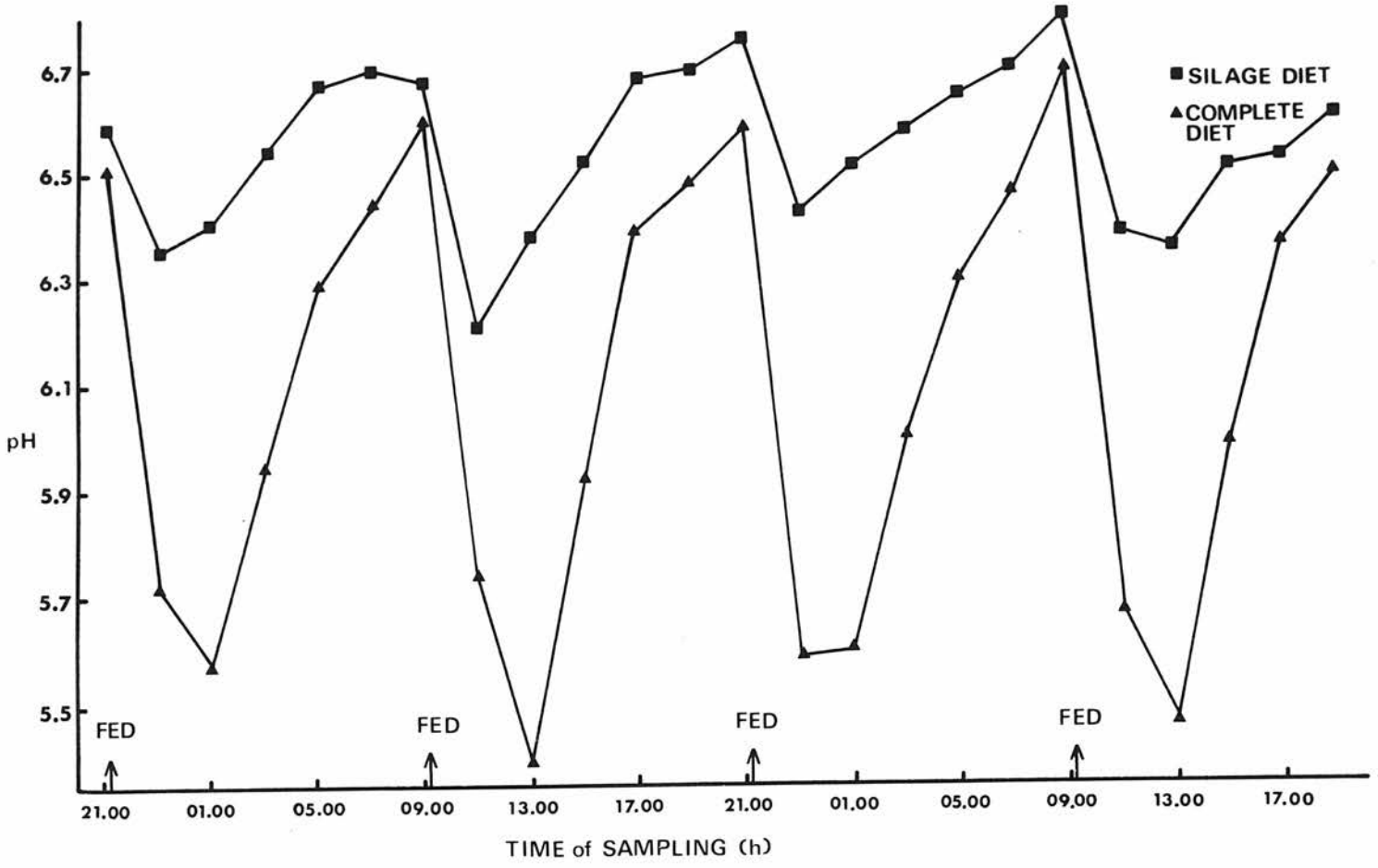

Fig. 7 CHANGES IN RUMINAL pH VALUES

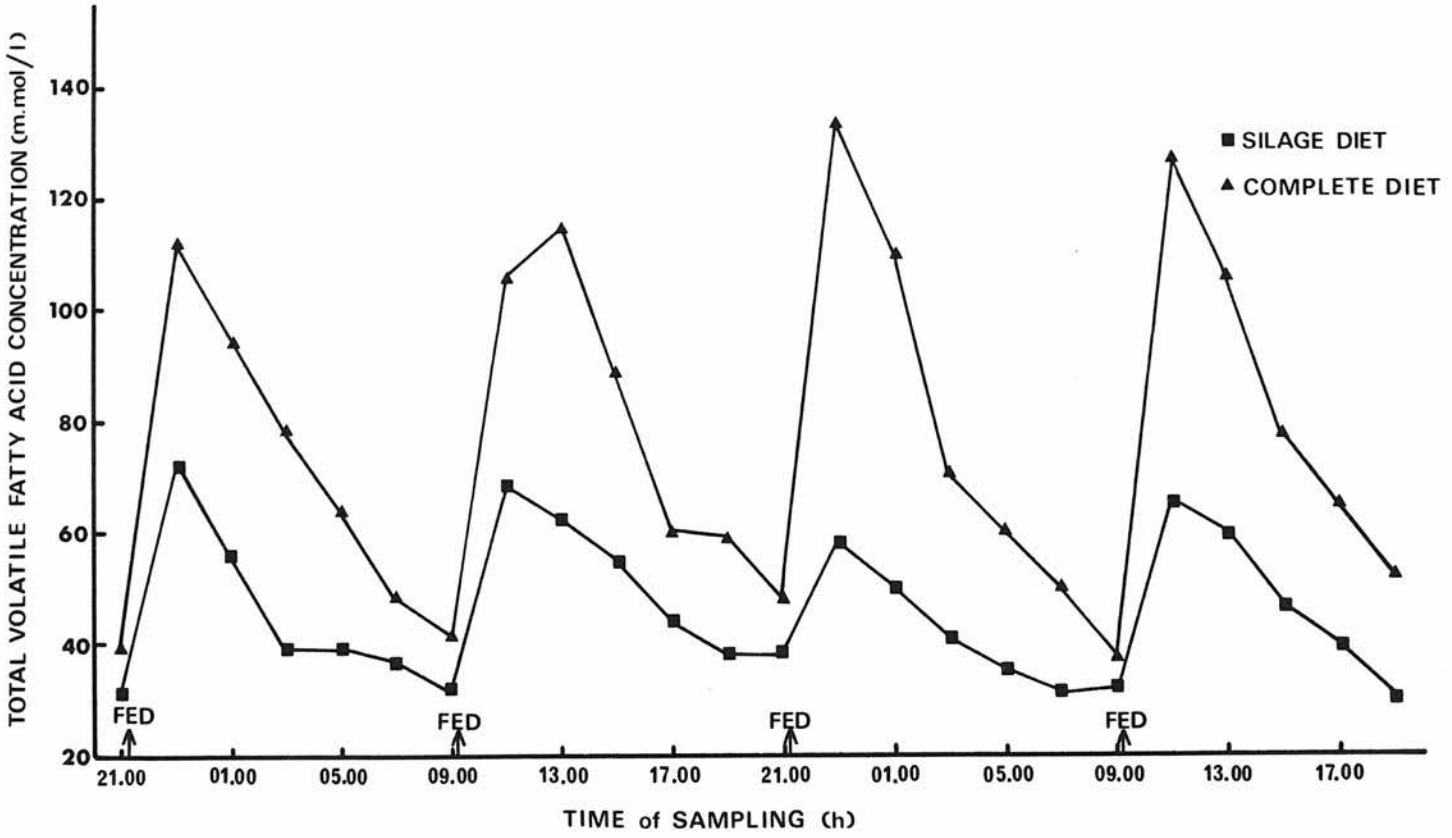

Fig. 8 CHANGES IN RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATION

sampled immediately before feeding and at two hourly intervals thereafter. After a changeover period intakes of dry matter were recorded for a further fourteen days. During the final forty eight hours rumen contents were sampled immediately before feeding, at four one-hourly intervals and then at two hourly intervals to the next pre-feeding sample.

RESULTS

Intake

Mean daily dry matter intakes were 37.8 and 59.2 g/kg $W^{0.75}$ for the silage and the complete diet respectively mean daily intakes for each animal are given in Appendix Table 1.

Rumen Characteristics

pH and TVFA Concentration: The results of the analyses carried out on the rumen liquor samples from individual sheep are given in Appendix Tables 2 to 13. Mean pH values of the rumen contents for both diets over the forty-eight hour sampling period are shown in Fig. 7. A regular pattern was obtained over each twelve hour sampling period and this pattern was repeated during the forty-eight hours. Maximum values occurred at the pre-feeding sampling and the pH fell sharply to a minimum at two or four hours after feeding; there then followed a gradual rise to the next pre-feeding sample.

Fig. 8 shows the mean ruminal total volatile fatty acid (TVFA) concentrations for the six sheep over the forty-eight hour sampling period. A regular pattern was obtained within each twelve hour period and this was repeated over the subsequent three feeding periods. Minimum concentration occurred at the pre-feeding sampling, there was a sharp increase in concentration to a maximum at two hours post-feeding with the silage diet and at two or four hours with the complete diet. The pattern

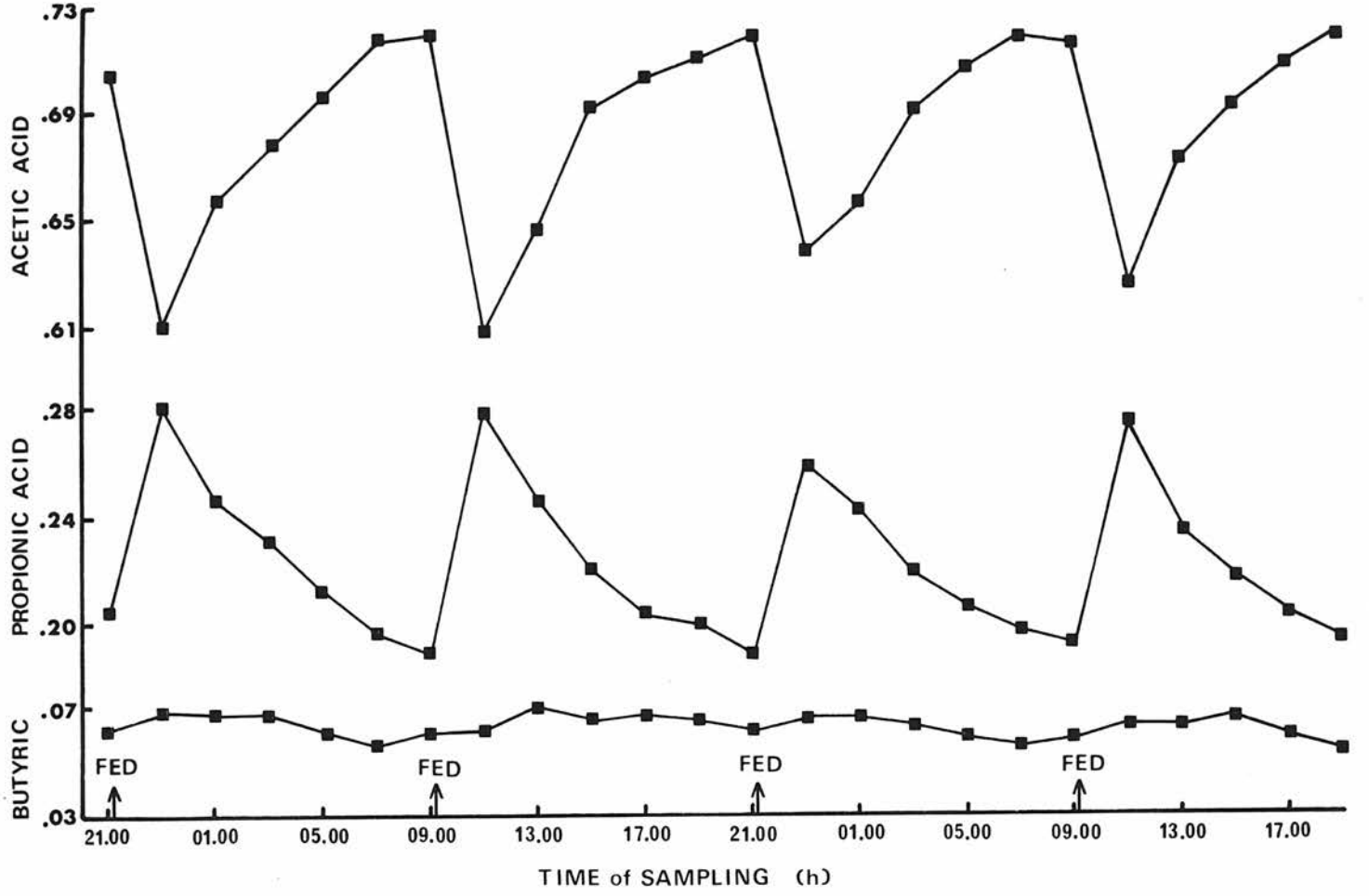

Fig. 9 CHANGES IN RUMINAL MOLAR PROPORTIONS OF ACETIC, PROPIONIC AND BUTYRIC ACID WITH SILAGE DIET

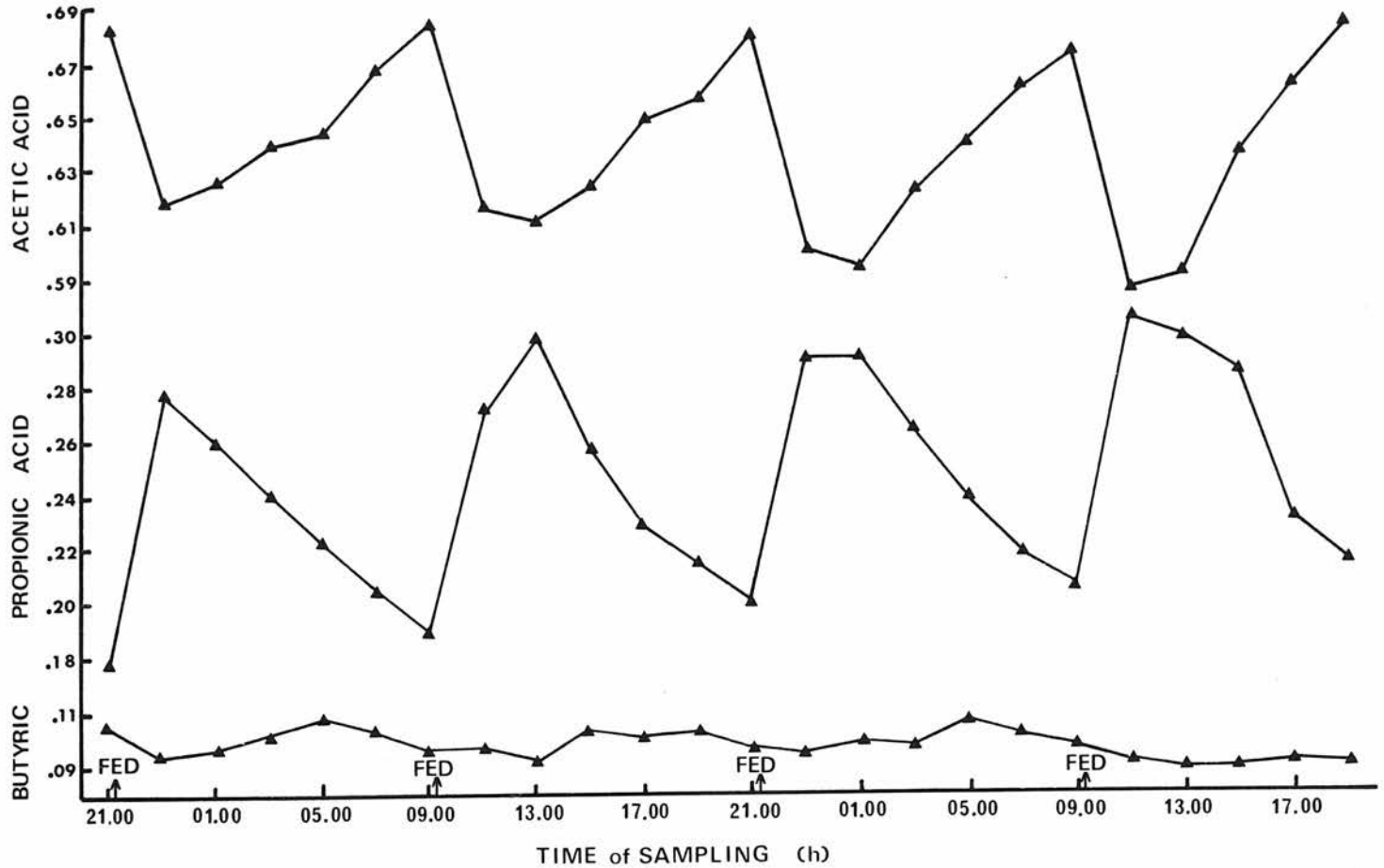

Fig. 10 CHANGES IN RUMINAL MOLAR PROPORTIONS OF ACETIC, PROPIONIC AND BUTYRIC ACIDS WITH COMPLETE DIET

of change of TVFA concentration during the forty-eight hour sampling reflected that for pH.

Volatile Fatty Acids: The individual acids detected in the rumen contents included acetic, propionic, n- and iso-butyric, n- and iso-valeric and hexoic. The range of acids was the same for both diets. Their concentration varied throughout the sampling period and the hexoic acid was generally present in trace quantities only.

Fig. 9 and 10 show the molar proportions of the three main rumen volatile fatty acids during four twelve-hour periods of sequential sampling following the ingestion of food. Acetic, propionic and butyric acid curves show a regular and repeatable twelve hour pattern with both diets.

The maximum molar proportion of acetic acid in the rumen occurred at pre-feeding. The proportion decreased sharply to a minimum value at two hours post-feeding with the silage diet and at two and four hours post-feeding with the complete diet. Minimum values of propionic acid for both diets were at pre-feeding.

The range of the molar proportions of n-butyric acid was very small. The widest range occurred with the silage and extended from 0.070, four hours after feeding, to 0.054, ten hours after feeding. With the complete diet the greatest range between maximum and minimum values was 0.020.

The sum of the molar proportions of the minor volatile fatty acids of the rumen during the forty-eight hour sampling period are shown in Fig. 11. Within each twelve hour period there is a regular pattern and this is repeated for the four periods with both diets. For the silage diet maximum values occurred at two hours post-feeding, except in the second period when it was at four hours post feeding.

Ammonia Concentration: Mean ruminal ammonia concentrations over the forty-eight hour sampling period are shown in Fig. 12 and 13. A regular pattern emerged within

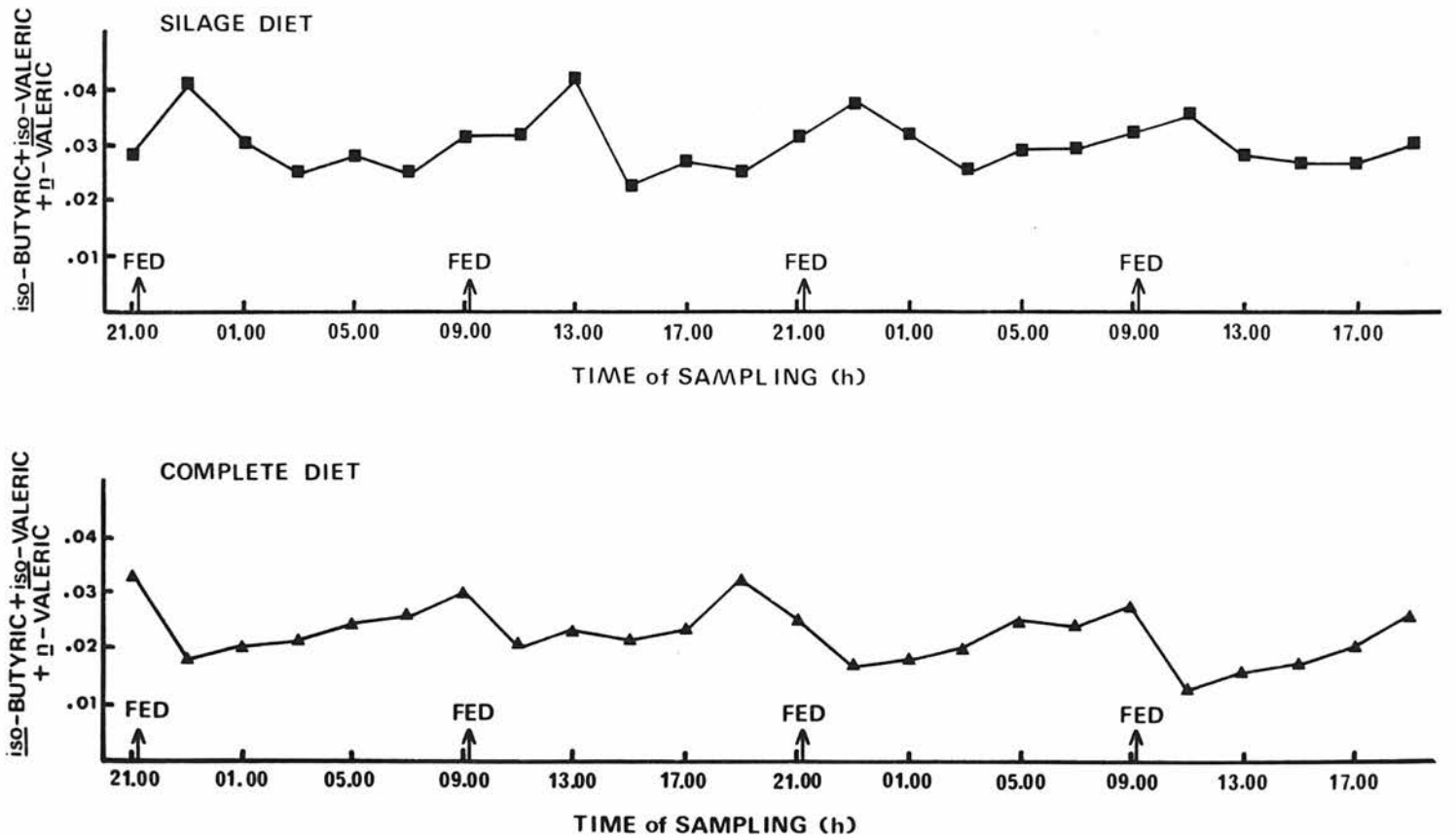

Fig. 11 CHANGES IN RUMINAL MOLAR PROPORTIONS OF iso - BUTYRIC plus iso - VALERIC plus n - VALERIC ACID

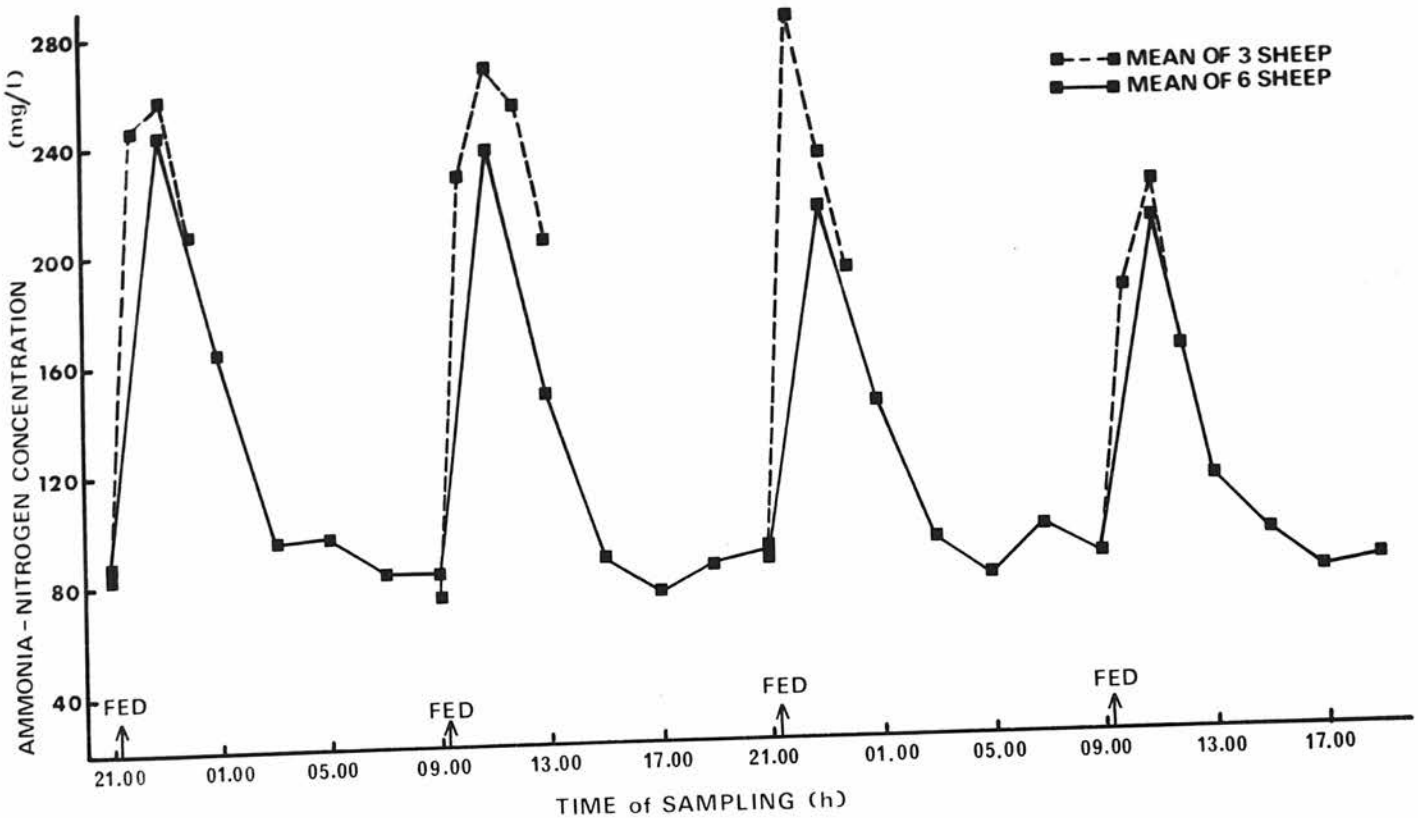

Fig.12 CHANGES IN RUMINAL AMMONIA CONCENTRATION WITH SILAGE DIET

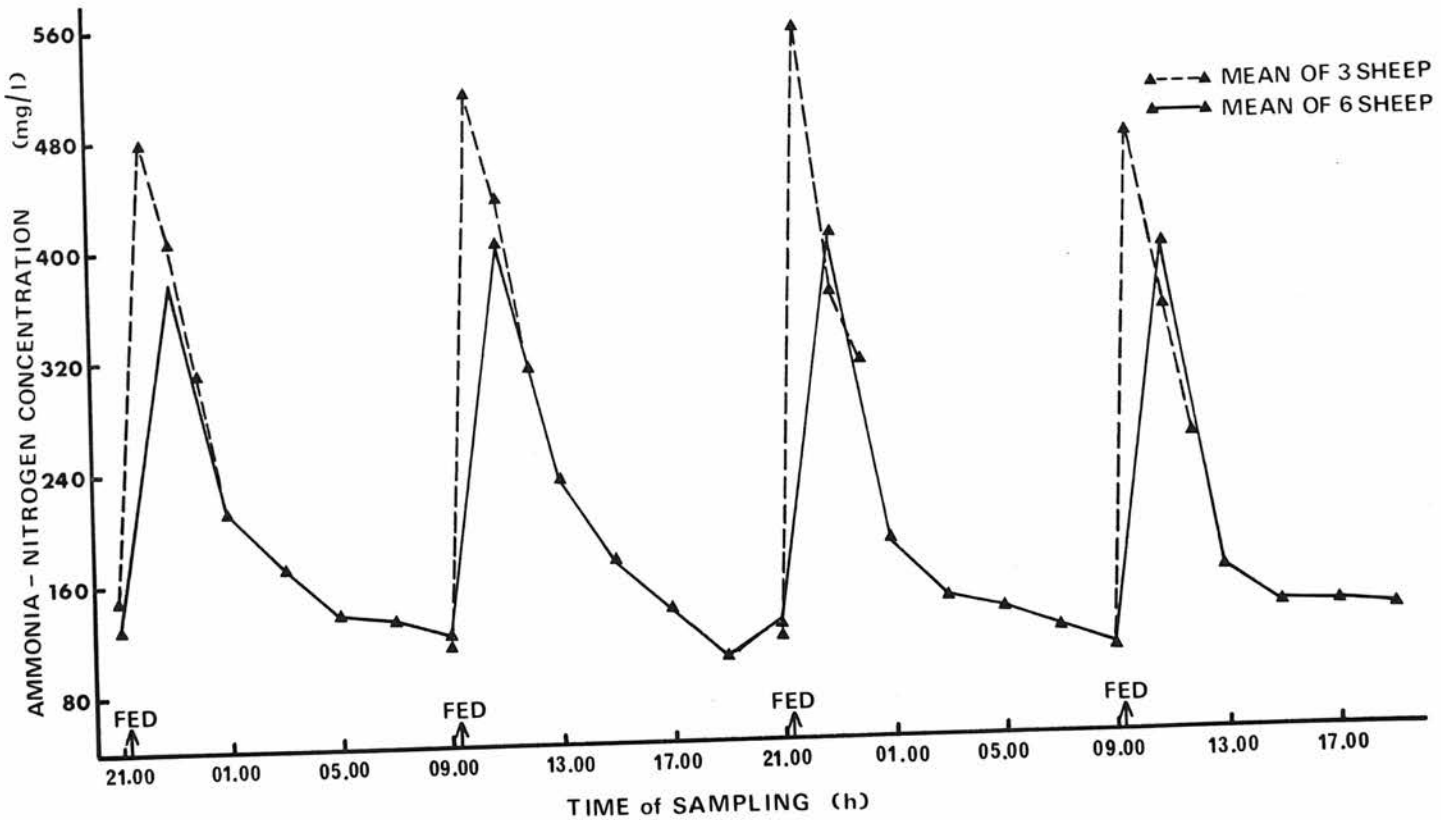

Fig. 13 CHANGES IN RUMINAL AMMONIA CONCENTRATION WITH COMPLETE DIET

each twelve hour period with both diets. Minimum values occurred at pre-feeding. There was a sharp increase to maximum concentration at two hours post-feeding, followed by a decrease to almost minimum concentration at six hours post-feeding. However, when the rumen contents were sampled more frequently, maximum concentration was at one hour post-feeding with the complete diet and at one or two hours with the silage diet. These were mean values from three animals only. The same twelve hour pattern following ingestion of food was repeated during the forty-eight hour sampling period.

The curves shown in Fig. 14 to 21 illustrate the changes in rumen pH, TVFA concentration, molar proportions of acetic and propionic acids and ammonia concentration for individual animals. Each point is the mean value of four determinations carried out on rumen samples taken at the same time on four different occasions.

With each parameter the pattern of change in individual animals showed considerable variations.

DISCUSSION

Several investigators, Schambye and Phillipson (1949) with a hay plus meal diet, Moir and Somers (1957) with a wheaten lucerne chaff plus cubes, Balch and Rowland (1957) with hay and hay plus concentrate, and Briggs *et al.* (1957) with wheaten and lucerne chaff, alone or supplemented with grain, or with grain plus a protein concentrate, have shown that fermentation activity within the rumen is minimal shortly before feeding and maximal shortly afterwards.

Bath and Rook (1963) have shown variations in the molar proportions of acetic, propionic and butyric acids in the rumen contents sampled at intervals following the ingestion of food. The fall in pH and rise in TVFA concentration at this time was generally accompanied by a decrease in the molar proportion of acetic acid and corresponding increases in the other two major acids as reported by Gray and Pilgrim

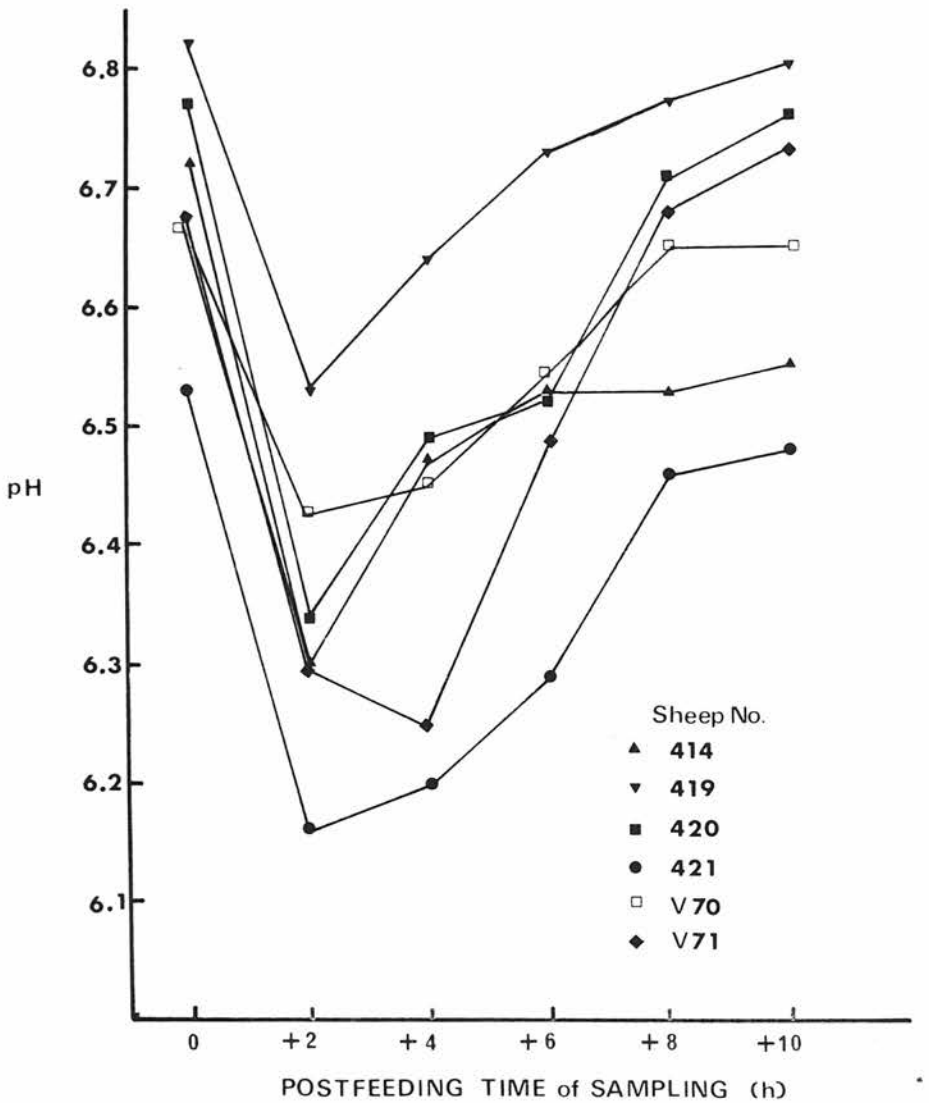

Fig. 14 RUMINAL pH VALUES FOR INDIVIDUAL SHEEP ON SILAGE DIET

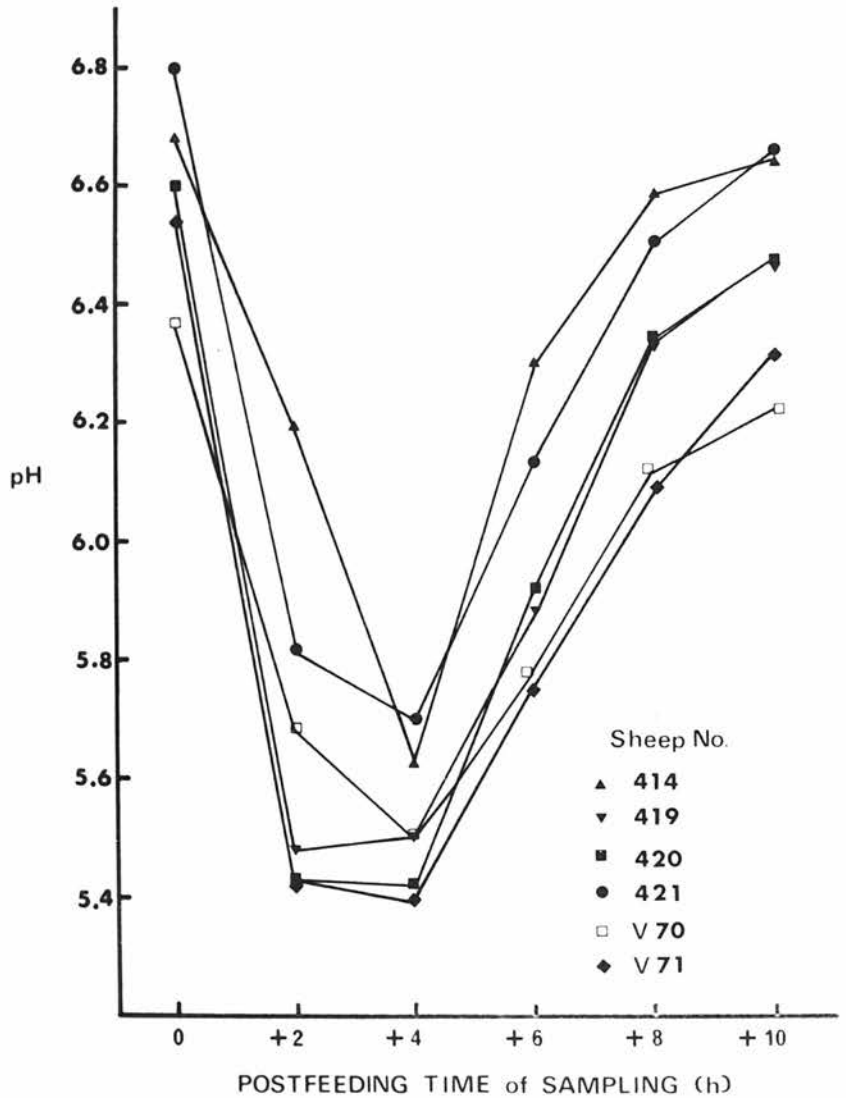

Fig. 15 RUMINAL pH VALUES FOR SHEEP ON COMPLETE DIET

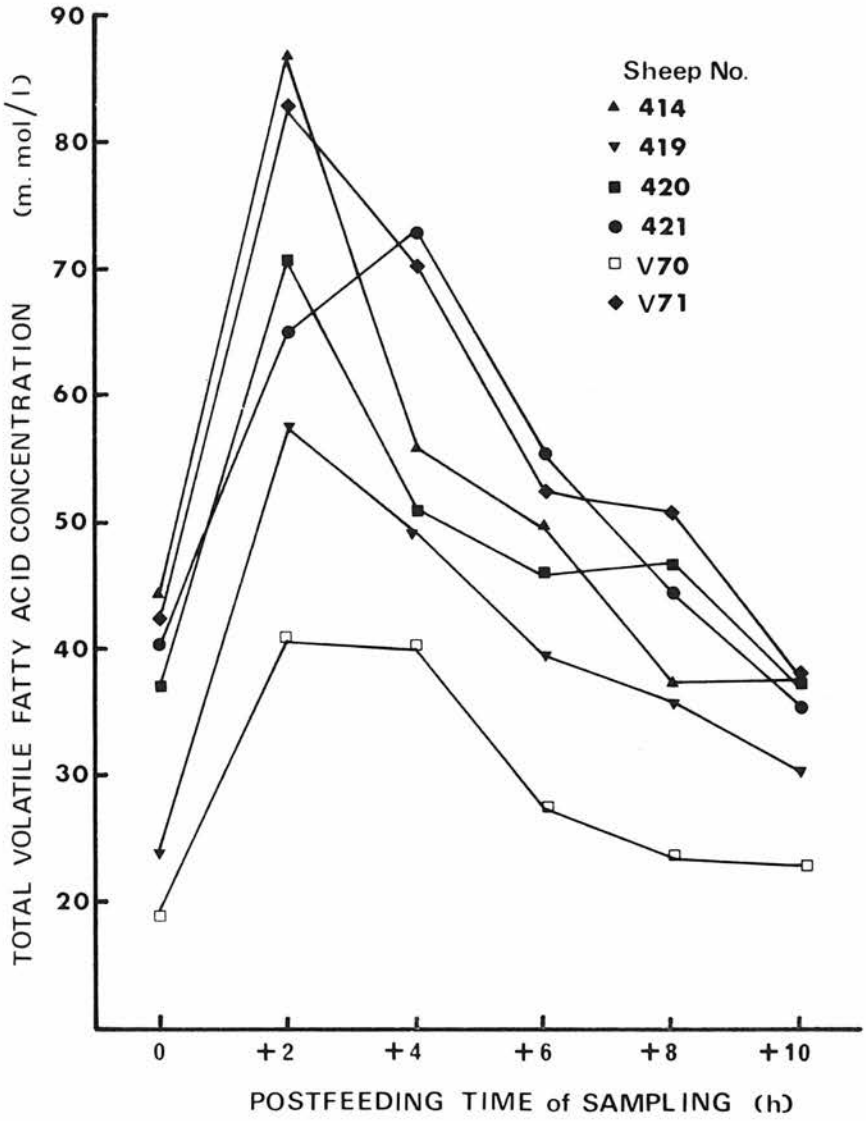

Fig. 16 RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATION FOR INDIVIDUAL SHEEP ON SILAGE DIET

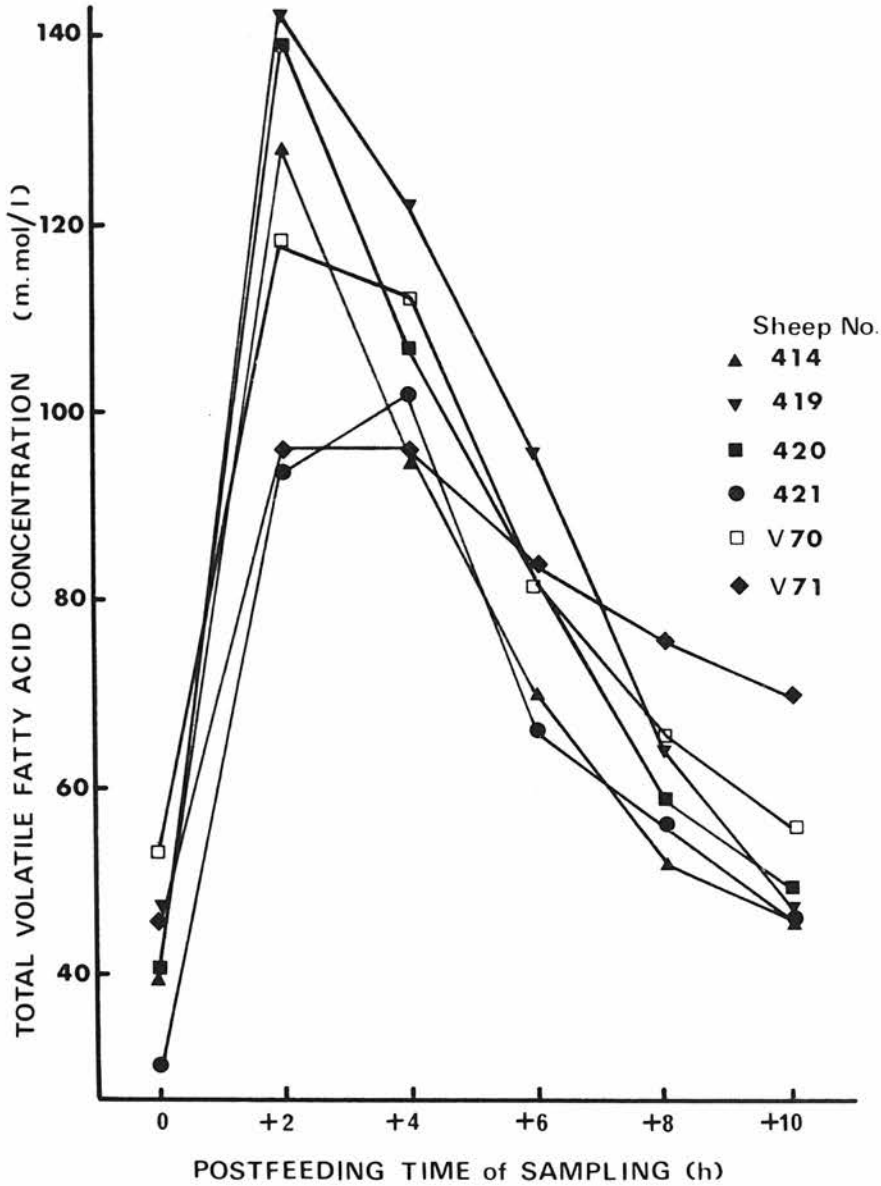

Fig. 17 RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATION FOR INDIVIDUAL SHEEP ON COMPLETE DIET

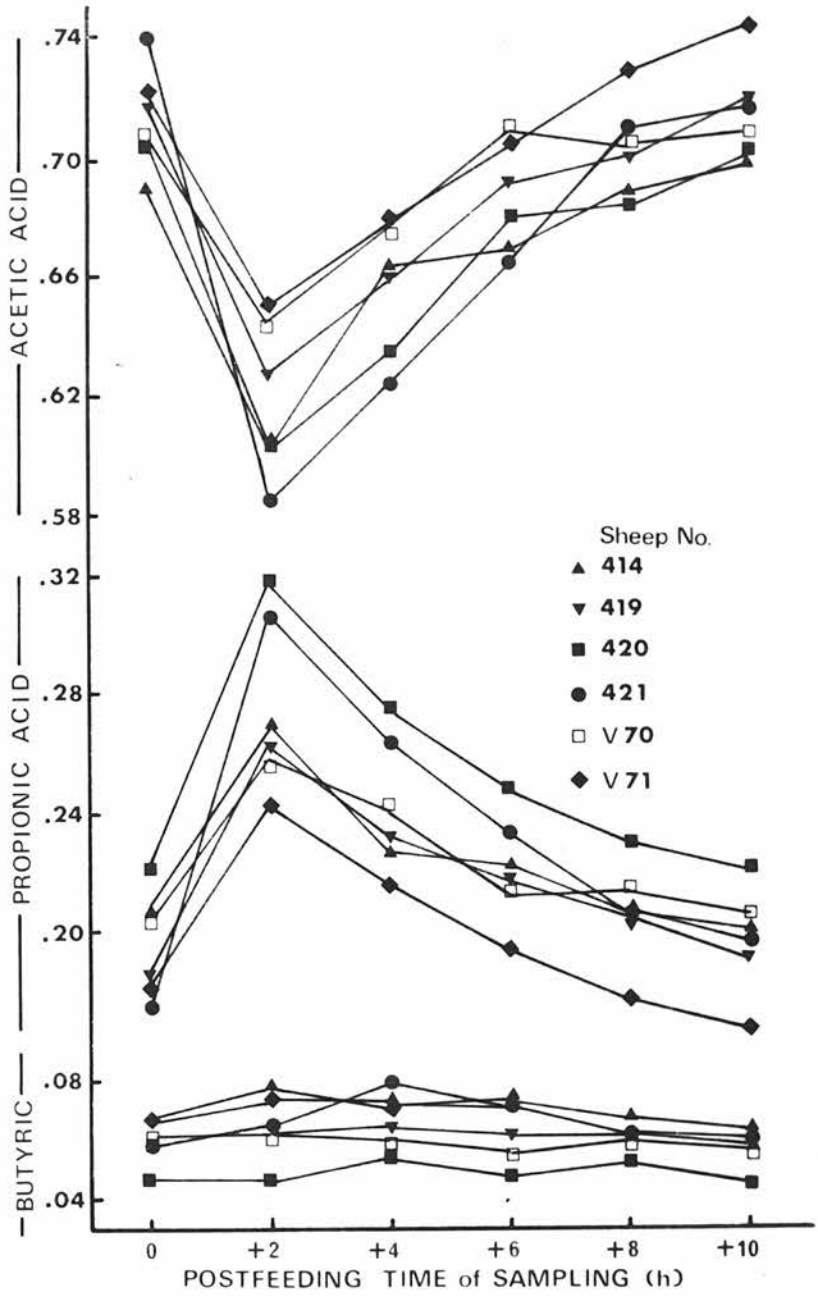

Fig. 18 RUMINAL MOLAR PROPORTIONS OF ACETIC, PROPIONIC AND BUTYRIC ACIDS FOR INDIVIDUAL SHEEP ON SILAGE DIET

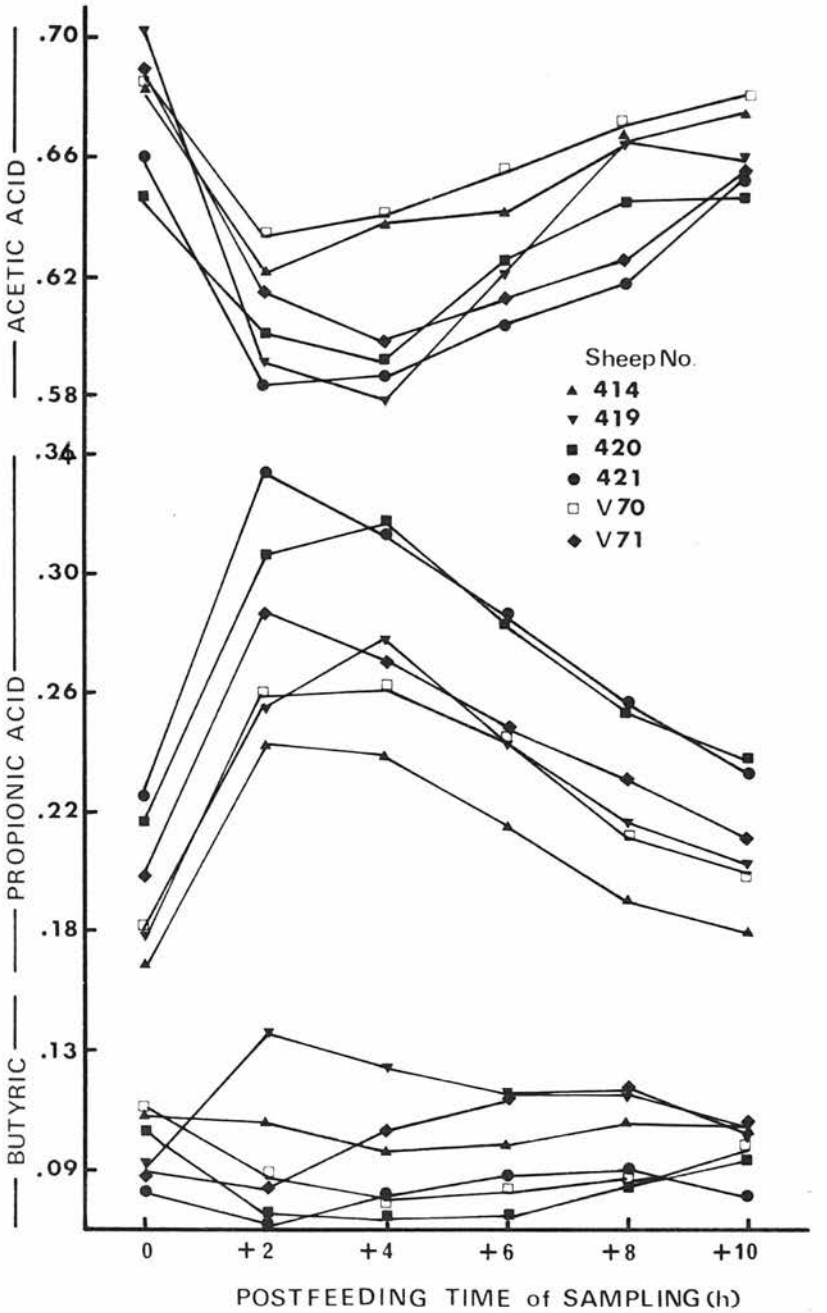

Fig. 19 RUMINAL MOLAR PROPORTIONS OF ACETIC, PROPIONIC AND BUTYRIC ACIDS FOR INDIVIDUAL SHEEP ON COMPLETE DIET

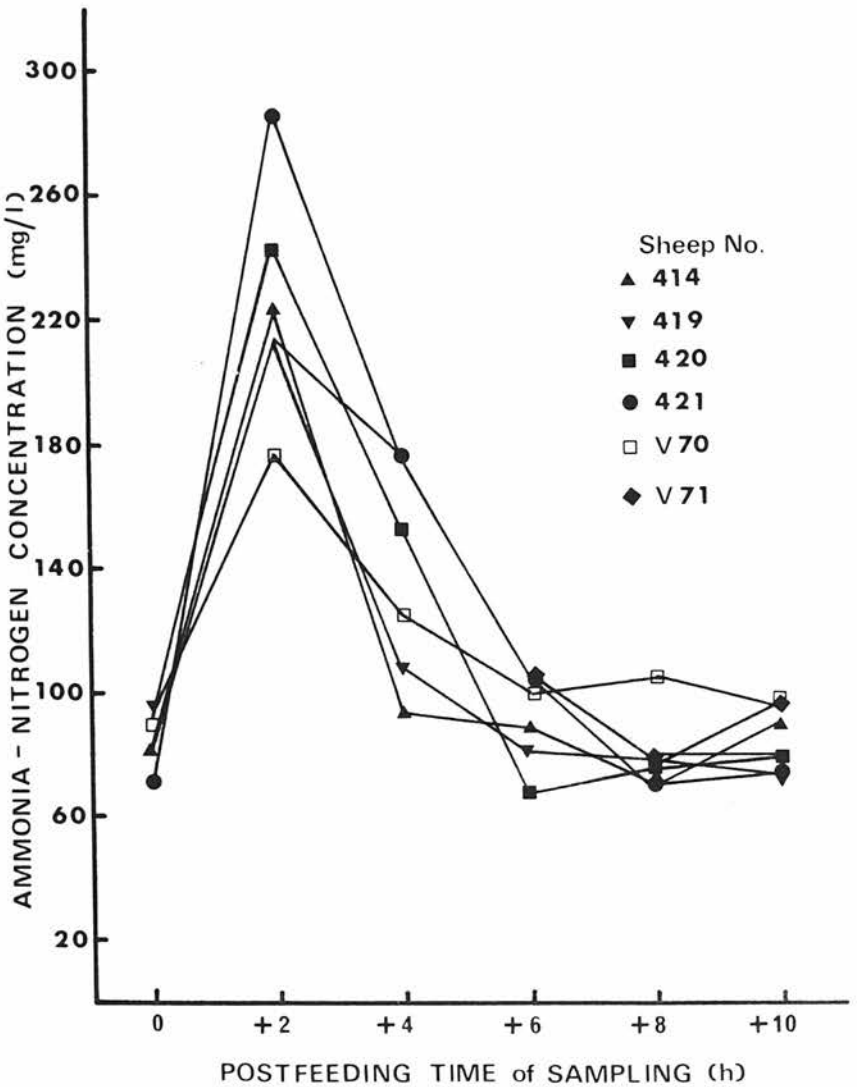

Fig. 20 RUMINAL AMMONIA CONCENTRATION FOR INDIVIDUAL SHEEP ON SILAGE DIET

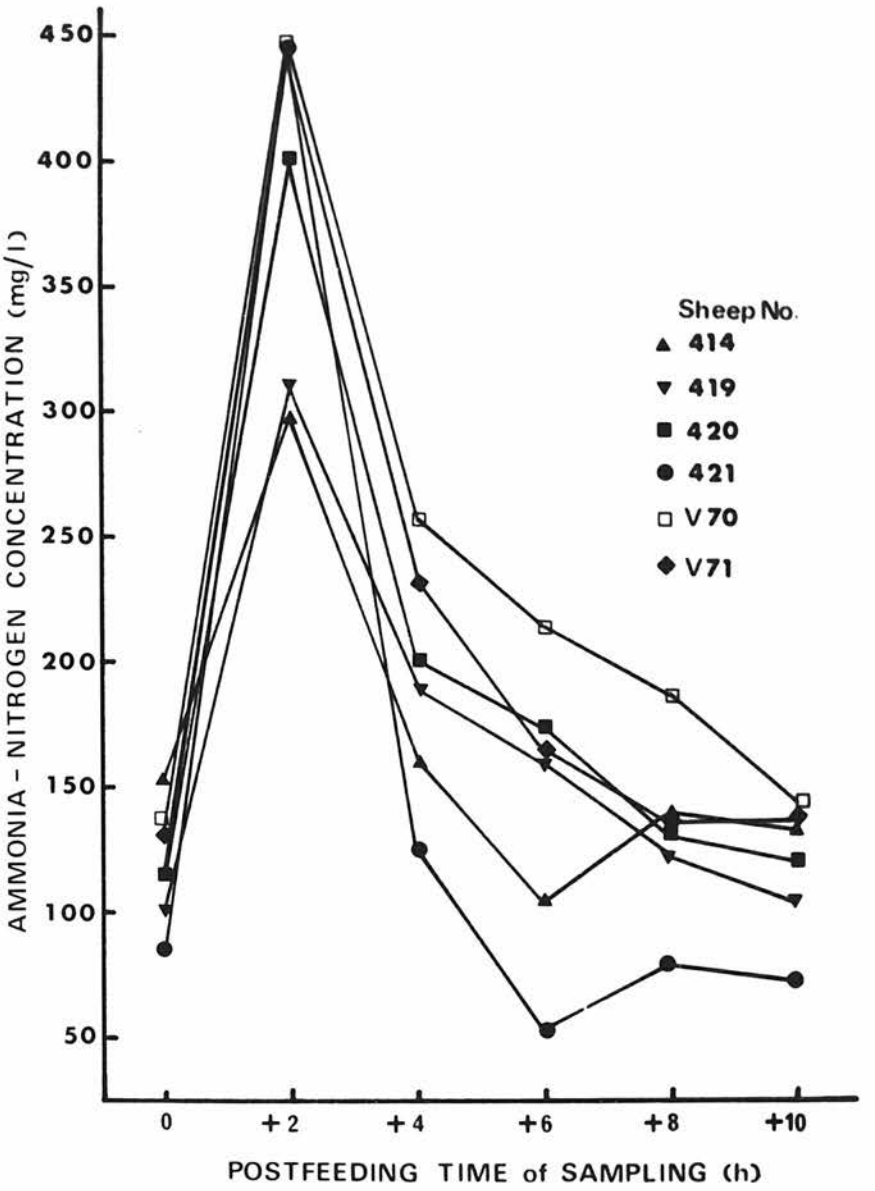

Fig. 21 RUMINAL AMMONIA CONCENTRATION FOR INDIVIDUAL SHEEP ON COMPLETE DIET

(1951) and Reid et al. (1957).

Shaw (1961), however, considered the variation in the molar proportions of the individual rumen acids throughout the feeding cycle to be small and that a single sample drawn at any one time during a feeding cycle was adequate to characterise the diet. Armstrong (1961) with a diet of dried grass confirmed Shaw's findings but with diets containing concentrates found some variation. Schambye and Phillipson (1949) showed a variation with time of sampling but no regular pattern. Moir and Somers (1957) with the diet of chaff plus cubes, Annison et al. (1959) with diets of hay alone, hay supplemented with casein or hay supplemented with flaked maize, and El Shazly (1952) with silage and grass diets reported a regular pattern of rumen ammonia concentration during the feeding cycle. Chalmers (1961) stressed that when rumen ammonia concentrations are determined at intervals, the curve obtained follows the intake of nitrogen. It would appear, therefore, that in using rumen ammonia concentrations for evaluating dietary nitrogen sources, the feeding regime has to be standardised and concentration curves, rather than single values, must be used.

The results of the present investigation show that the rumen patterns of pH, TVFA concentration, the molar proportions of acetic, propionic and butyric acids and ammonia concentration are similar for the silage and the complete diet and confirm the general post-feeding changes shown by earlier workers. For the latter diet the pattern of change was similar to that recorded by Gray and Pilgrim (1951) for a hay diet and Balch and Rowland (1957) for a hay plus concentrate diet. The changes in rumen TVFA and ammonia concentration are similar to those recorded by Williams and Christian (1959) for silage diets. Fenner et al. (1970) have reported statistically significant differences in the molar proportions of acetic and butyric acids between hourly samples of rumen contents when diets of hay, silage or hay plus silage were fed.

Bath and Rook (1963) stressed the need to sample rumen contents at frequent intervals but they quote only average values of pH, TVFA concentration and the molar proportions of the individual acids. The present data confirm the need for frequent sampling but illustrate too the inadequacy of a mean figure in characterising rumen fermentation, as do those of Fenner et al. (1967). More recently Du Plessis et al. (1969) stated that there was no single best time for estimating rumen characteristics and Steger et al. (1970) have confirmed the need to know the curve pattern of rumen fermentation.

Williams and Christian (1956) found little difference between days in the rumen microbial end products of grazing sheep. With animals fed indoors Bath and Rook (1963) reported day to day variations within two cows in rumen pH with values of 6.26 to 6.00 and 6.15 to 6.01, and in TVFA concentration from 86.7 to 83.4 and 100.2 to 90.6 m mol/l over four days. Fenner et al. (1967) reported highly significant differences in daily rumen pH values and in the proportion of n-butyric acid between daily measurements at three hours post-feeding when the diet was hay. The results of the present investigation tend to confirm day to day differences between the values for rumen characteristics in individual animals.

The lack of variation between the two day and the two night rumen fermentation patterns based on mean values for animals on the silage and the complete diet is in agreement with the findings of Steger et al. (1970) who reported a variance ratio in the same animal from day to day to 0.2 units of pH, 18.4 m mol/l of TVFA and 40 mg/l of ammonia nitrogen concentration.

The repeatability of the twelve hour rumen fermentation patterns following the ingestion of the silage or the complete diet during the forty-eight hour sampling period confirm the results of Balch and Rowland (1957) and Stewart et al. (1958) who found a similarity in rumen fermentation patterns after the morning and the evening feeding.

Williams and Christian (1956) reported a variation between grazing sheep when rumen pH, TVFA and ammonia concentrations were measured. Balch and Rowland (1957) and Bath and Rook (1961, 1963 and 1965) reported differences between rumen pH values, TVFA concentration and the molar proportions of acetic, propionic and butyric acids when hay plus concentrates were fed to cows. Chou and Walker (1964) showed differences in rumen pH and ammonia concentration between sheep fed diets of different starches. Chalmers (1961, 1963) showed differences between sheep in maximum and minimum concentrations when the rumen ammonia patterns were examined. Steger et al. (1970) for six cows on the same diet reported the molar proportions of acetic acid in the rumen to range from 0.740 to 0.781, of propionic acid from 0.154 to 0.186 and of butyric acid from 0.059 to 0.078. The difference between sheep, of approximately 0.05 units, in the molar proportion of acetic acid when either experimental diet was fed in the present investigation was similar to that for Steger's cows. The difference between sheep in the molar proportions of propionic and butyric acids, which were as much as 0.090 and 0.065 respectively, were much greater than those reported by Steger but no as great as those of 0.228 and 0.151 quoted by Ishaque et al. (1971). The latter workers claimed that sheep could be grouped according to the type of rumen fermentation. They showed differences between sheep in the pH, and concentrations of ammonia and TVFA in the rumen, when the sheep were given a standard ration. pH values ranged from 5.53 to 6.03, ammonia nitrogen values from 111 to 270 mg/l and TVFA concentrations from 73.2 to 132 m mol/l for four sheep fed at hourly intervals. In the present investigation maximum differences for individual sheep, over the between-feeding period, were 6.18 to 6.52 for pH, 175 to 280 mg/l for ammonia nitrogen concentration and 40 to 85 m mol/l for TVFA concentration.

Du Plessis et al. (1969) in experiments to investigate the number of sheep required for a metabolism trial measured the pH, TVFA and ammonia concentrations in

the rumen at thirteen two hour intervals and concluded that since observations on the same sheep in successive experiments were not significantly correlated the same sheep could be studied repeatedly.

CONCLUSIONS

The between animal differences in rumen characteristics demonstrate the need for animal replication and the desirability of a cross over technique so that each individual animal's reaction to a particular diet is included in the final analysis.

The differences in the rumen parameters with time after feeding require that comparison be made on the basis of curve patterns of rumen fermentation rather than mean values for several samples taken over the period, or values for single samples taken at a fixed time after feeding.

The similarity of curve pattern between days and between nights and between days and nights allows sampling during the day only. In order to allow for any day to day variation, however small, samples should be taken on two consecutive days and comparable samples (time after feeding) bulked for analysis.

V

EXPERIMENT 2

THE EFFECT ON RUMEN FERMENTATION CHARACTERISTICS OF CHANGES IN THE METHOD OF FEEDING A SILAGE DIET

INTRODUCTION

The previous experiment showed that a series of comparable and repeatable rumen fermentation curves could be obtained, with both a silage diet and a complete diet, when the time allowed for feeding was restricted to two hours at twelve hour intervals. The effects on rumen fermentation of modifying this arbitrary feeding method was investigated by comparing it with a regime in which silage was fed at twelve hour intervals with free access between meals, and another in which silage was given six times per day at four hour intervals with access time limited to forty minutes.

EXPERIMENTAL

Nine sheep were used in a cross-over design with three sheep on each treatment during each period. Sheep were allocated at random to different treatments and the design was balanced for residual effects. The design is shown in Table 10.

Table 10

Cross-over Design for Three Treatments and Nine Sheep

Sheep No. / Period	1 (437)	2 (409)	3 (449)	4 (88)	5 (414)	6 (435)	7 (680)	8 (434)	9 (447)
I	A	B	C	A	B	C	A	B	C
II	B	C	A	C	A	B	B	C	A
III	C	A	B	B	C	A	C	A	B

The treatments were:

A. Sheep were fed two meals of 650 g of silage dry matter at 09.00 and 21.00 h.

and residues were removed at 20.50 and 08.50 respectively.

B. Sheep were fed two meals of 650 g of silage dry matter at 09.00 and at 21.00 h. and residues were removed at 11.00 and 23.00 h. respectively.

C. Sheep were fed six meals of 217 g of silage dry matter at 09.00, 13.00, 17.00, 21.00, 01.00 and 05.00 h. and residues were removed at 09.40, 13.40, 17.40, 21.40, 01.40 and 05.40 h. respectively.

The silage was made from Italian ryegrass with a dry matter of 184 g/kg, cut on 31st May and wilted for twenty four hours during which there was evening and overnight rain. The grass was lifted with a double chop forage harvester and ensiled in two-tonne plastic silos. Two silos were opened after 77 days, the silage mixed, bagged and stored for feeding. The toluene dry matter of the silage was 217 g/kg and the pH 3.95. The silage composition is given in Table 11.

Table 11

Composition of Silage (g/kg dry matter)

Total nitrogen	29
Water soluble nitrogen	7.4
Volatile nitrogen	1.0
MAD - fibre	369
Ash	98
Water soluble carbohydrate	42
Lactic acid	60
Succinic acid	2.9
Acetic acid	15.8
Propionic acid	0.6
Butyric acid	3.2

The normal procedure for sampling rumen contents was adopted for groups A and B while group C animals were sampled every hour throughout the twelve hour cycle.

RESULTS AND DISCUSSION

Nutritive Value

Intake: The mean dry matter intakes of the silage for the three treatments A, B and C were 47.6, 38.7 and 45.8 g/kg $W^{0.75}$ respectively. Analysis of variance showed significant differences between the twice daily feeding with limited access and the other two treatments. Mean daily intakes for individual sheep on each treatment are given in Appendix Table 14.

The increased intake with the increase in the frequency of feeding from two to six times daily agrees with the results for mixed roughage concentrate diets found by Campbell and Merilan (1961) for cows, and by Mohrman et al. (1959) for beef cattle, when the frequency was increased from twice to four times and from twice to six times daily respectively. Murdoch (1964) also found an increase from 949 to 1189 g in mean daily intake of silage by sheep, when the access time was increased from three to twenty-four hours. Hemminger and Kirchgessner (1972) obtained similar results when grass and maize silage plus hay was fed twice daily, three times daily and ad libitum to heifers. Mean daily intakes of silage dry matter were 5.98, 6.28 and 6.28 kg respectively. These authors attributed the higher intakes of once daily feeding with free access, and several times daily to the close resemblance of these regimes to the normal pattern of cows eating forage noted by Lewis and Johnson (1954) and Hanssen (1959). These results are not in accord with the findings of Gordon and Tribe (1952) who found diminished appetite in sheep when the frequency of feeding a mixed diet of roughage and concentrate was increased. Hillier et al. (1968) showed intakes of a maize silage by growing steers fed six times and twice daily were similar at 15.4 and 16.1 kg daily. Clark and Keener (1962) fed a mixed roughage-concentrate diet in unrestricted amounts twenty-four times, ten times and twice daily and could show no difference in intake.

Digestibility: Dry matter digestibilities for the treatments A, B and C were 0.702, 0.717 and 0.715 respectively. Analysis of variance showed significant differences between twice daily feeding with free access and the other two treatments. Dry matter digestibilities for individual sheep are given in Appendix Table 14.

Blaxter et al. (1956a) found that by doubling the intake of long dried grass from 600 to 1200 g daily the apparent digestibility was reduced by 0.012 units. McDonald et al. (1973b) state that as a general rule doubling the maintenance ration reduces the digestibility of dry matter by 0.01 to 0.02 units. Waldo et al. (1966) reported an increase in digestibility of dry matter from 0.551 to 0.579 when the dry matter intake of alfalfa silage was increased from 11.4 to 12.6 g/kg body weight, but in a subsequent experiment found no difference in digestibility when alfalfa silage was fed at two levels of intake. It would appear that the 0.015 units of difference between treatments A and B could be partly explained by the difference in dry matter intake of 221 g but may also reflect a different pattern of intake as well.

The six times daily feeding, with restricted access, gave an improved digestibility of dry matter of the silage of 0.013 units compared with twice daily feeding, with free access. This is not accounted for by the difference in intake of 47 g. Zero, positive and negative responses in the digestibility of the diet to increased frequency of feeding have been reported. Moir and Somers (1957) obtained a significant increase in dry matter digestibility when sheep were fed concentrate cubes plus chaff twice or four times daily instead of once. Campbell and Merilan (1961) also reported increased digestibility of dry matter when cows were fed at increased frequency. Blaxter et al. (1956b) obtained no response in digestibility values when sheep were fed equal amounts of dried grass at twenty-four, twelve and six hourly intervals. Satter and Baumgardt (1962) with cattle on a hay diet, Rhodes and Woods (1962) with lambs fed a roughage diet, Faichney (1968) and

Hillier et al. (1968) reported similar conclusions. Rakes et al. (1957) reported that feeding ten times daily significantly depressed the digestibility of dry matter compared with twice daily feeding of mixed hay. Sutherland et al. (1963) indicated that the response varied with the diet, since continuous feeding compared with twice daily feeding gave increased digestibility of hay but not of dried grass. Certainly the present results would indicate beneficial effects on digestibility as a result of increased frequency of feeding.

Despite the difference in dry matter intake between treatments B and C there was no difference in dry matter digestibility. This may be due to a balancing of the opposing effects of intake and frequency of feeding.

Organic matter digestibility, nitrogen digestibility, the gross energy of the digestible organic matter and the metabolisable energy for the three treatments are shown in Table 12. The values for individual sheep are given in Appendix Table 14.

Table 12

Nutritional Characteristics of the Silage

	A (twice daily, free access)	B (twice daily, limited access)	C (six times daily, limited access)
Digestibility of organic matter	0.74	0.75	0.75
Digestibility of nitrogen	0.61	0.63	0.63
Metabolisable energy (MJ / kg)	11.1	11.1	11.3
Gross energy of digestible organic matter (MJ / kg)	20.0	19.7	20.0

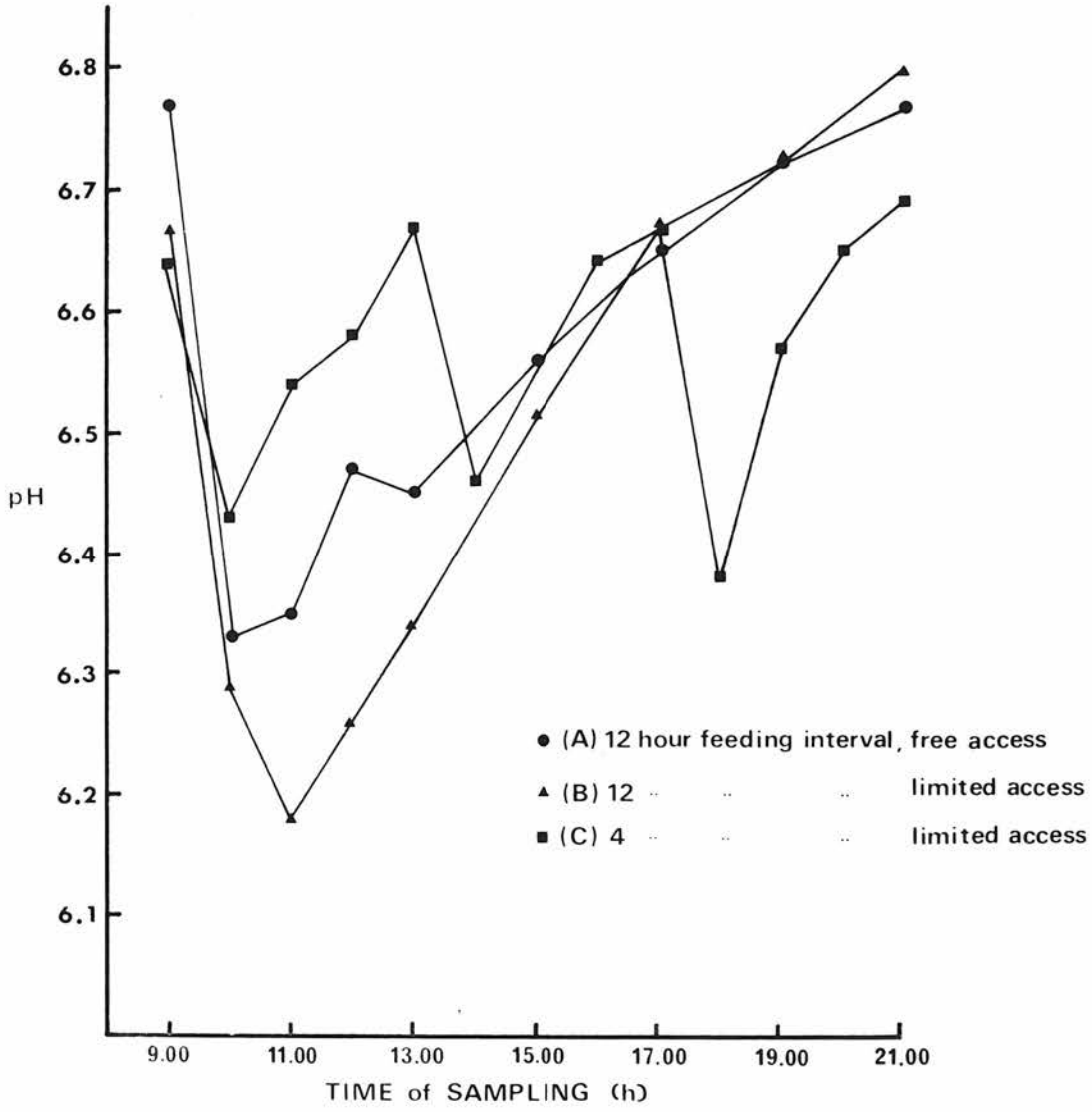

Fig. 22 RUMINAL pH VALUES

Analysis of variance gave no significant evidence for differences between the treatments for any of these values. Moir and Somers (1957) obtained significant increases in nitrogen digestibility, and Mohrman et al. (1959) obtained similar increases in digestibility of nitrogen and of energy by more frequent feeding. However, Rakes et al. (1957) found the digestibility of nitrogen was depressed when the frequency of feeding hay was increased, and McGuire et al. (1966) reported similar results.

The results of the present investigation are in agreement with the work of Graham (1967) who quoted organic matter digestibilities of 0.64 and 0.63, and crude protein digestibilities of 0.76 and 0.74 for a pelleted lucerne diet fed at three hour and twenty-four hour intervals respectively. Neither Rhodes and Woods (1962b) nor Faichney (1968) could find differences in nitrogen balance which could be attributed to differences in the frequency of feeding.

Rumen Characteristics

Rumen fermentation patterns of pH, ammonia and total volatile fatty acid (TVFA) concentrations, and the molar proportions of the rumen acids are based on mean values for nine sheep over two sampling cycles. The composition of rumen contents from the individual sheep on each treatment during the twelve hour feeding cycle are given in Appendix Tables 15 to 41.

pH and TVFA Concentration: pH values are shown in Fig. 22. The pattern between feeding intervals is similar for each treatment. Maximum values which were at pre-feeding were similar for the two treatments involving twice daily feeding but the minimum value for the treatment allowing limited access was lower and occurred later than for the other. Feeding six times daily reduced the range of variation of pH over the twelve hour period with values of 6.38 to 6.69 compared with 6.33 to 6.77

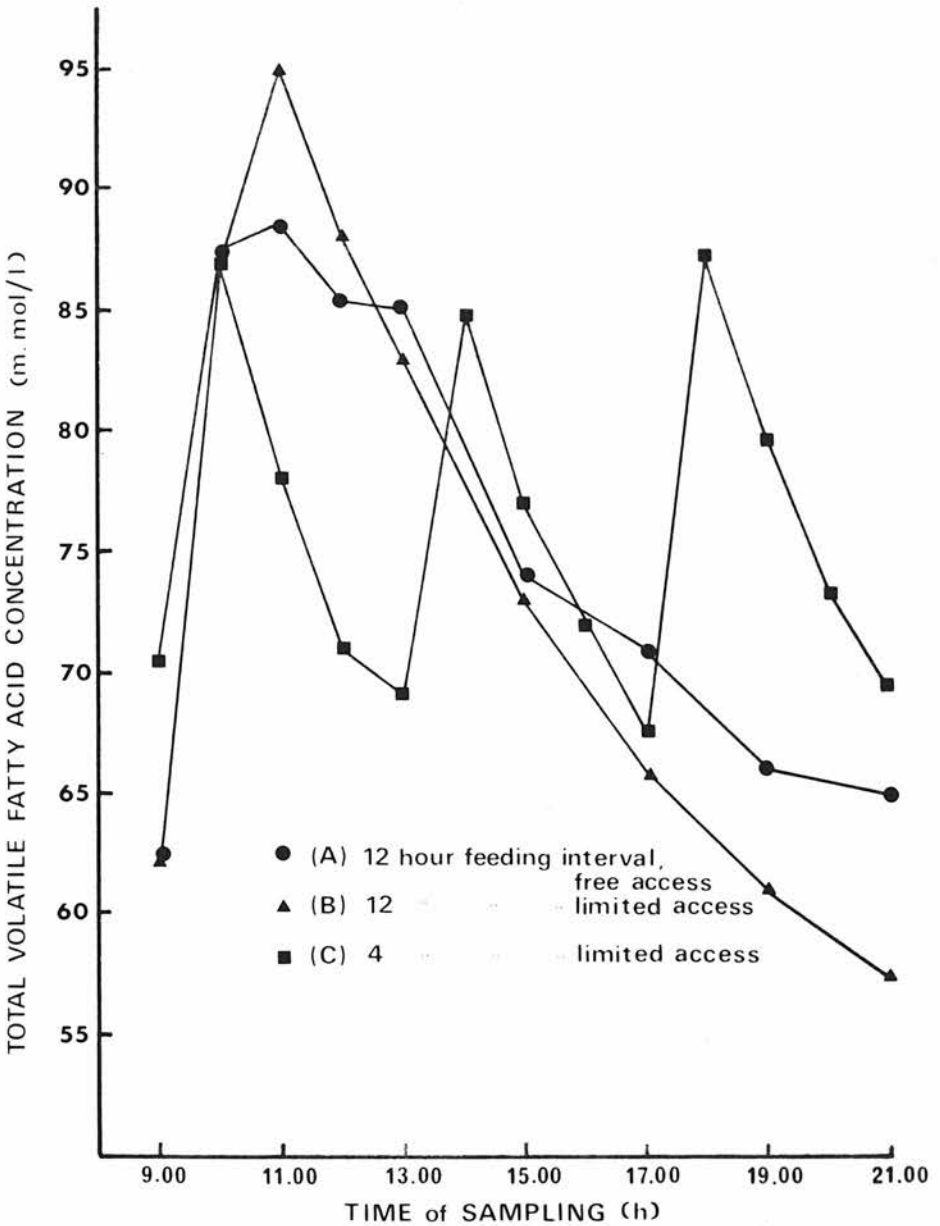

Fig. 23 RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATIONS

for twice daily with free access treatment, and 6.18 to 6.80 for the twice daily limited access treatment. Moir and Somers (1957) reported similar results for rumen samples taken from animals fed at varying frequencies. Bath and Rook (1963) for two cows fed hay once, twice or four times daily reported the range of values of 5.92 to 6.77, 6.24 to 6.70 and 6.29 to 6.56 respectively. With a diet of hay plus concentrate, the range of values was narrower with the more frequent feeding. The mean minimum values were lower with the six hourly compared with the twelve hourly feeding interval.

Rakes et al. (1961) found that the average pH did not differ significantly between once and eight times daily feeding, but reported that there was less fluctuation between maximum and minimum pH values with increased frequency of feeding. Satter and Baumgardt (1962) and Faichney (1968) reported similar findings.

Fig. 23 shows the rumen TVFA concentration patterns with the three treatments. Maximum values are lower and the decline from maximum is much slower for the treatment involving twice daily feeding with free rather than limited access. Comparison of the twice with six times daily feeding, both with limited access, shows a very similar pattern of rumen TVFA concentration, but with the four hour interval the pattern is condensed with a reduction in maximum and increase in minimum values.

Knox and Ward (1961) reported significant increases in TVFA concentration when the frequency of feeding a mixed barley plus hay diet was increased from twice to eight times daily and, with the latter, the pattern of concentration was less definite. Moir and Somers (1957) found little change in TVFA concentration at maximum levels when the diet was fed once, twice and four times daily, and Bath and Rook (1963) did not find differences in mean values when the feeding frequency was similarly increased. Putnam et al. (1961) found no apparent relationship between frequency of feeding and volatile fatty acid concentration.

The range of rumen TVFA concentration in the present investigation of 69 to

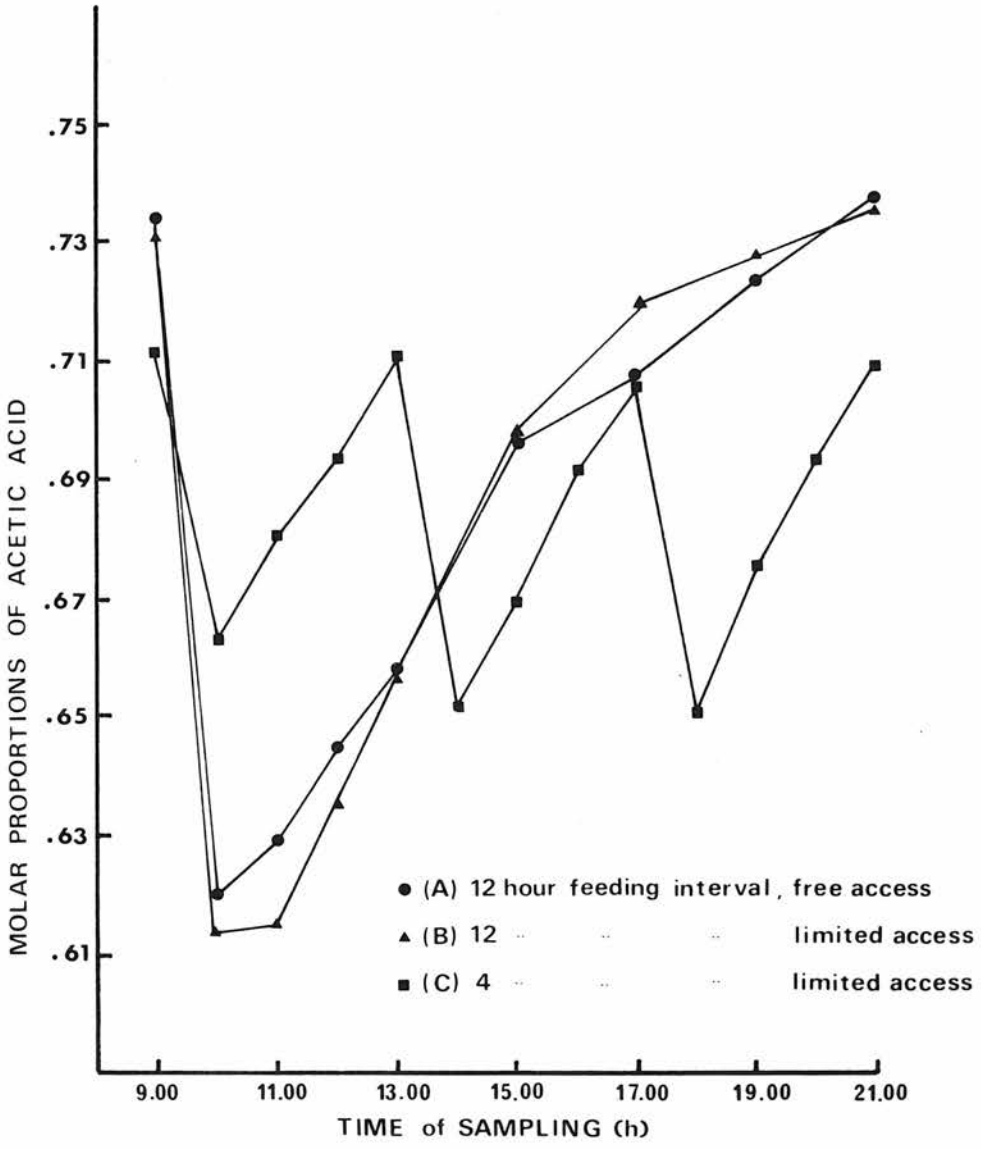

Fig. 24 RUMINAL MOLAR PROPORTIONS OF ACETIC ACID

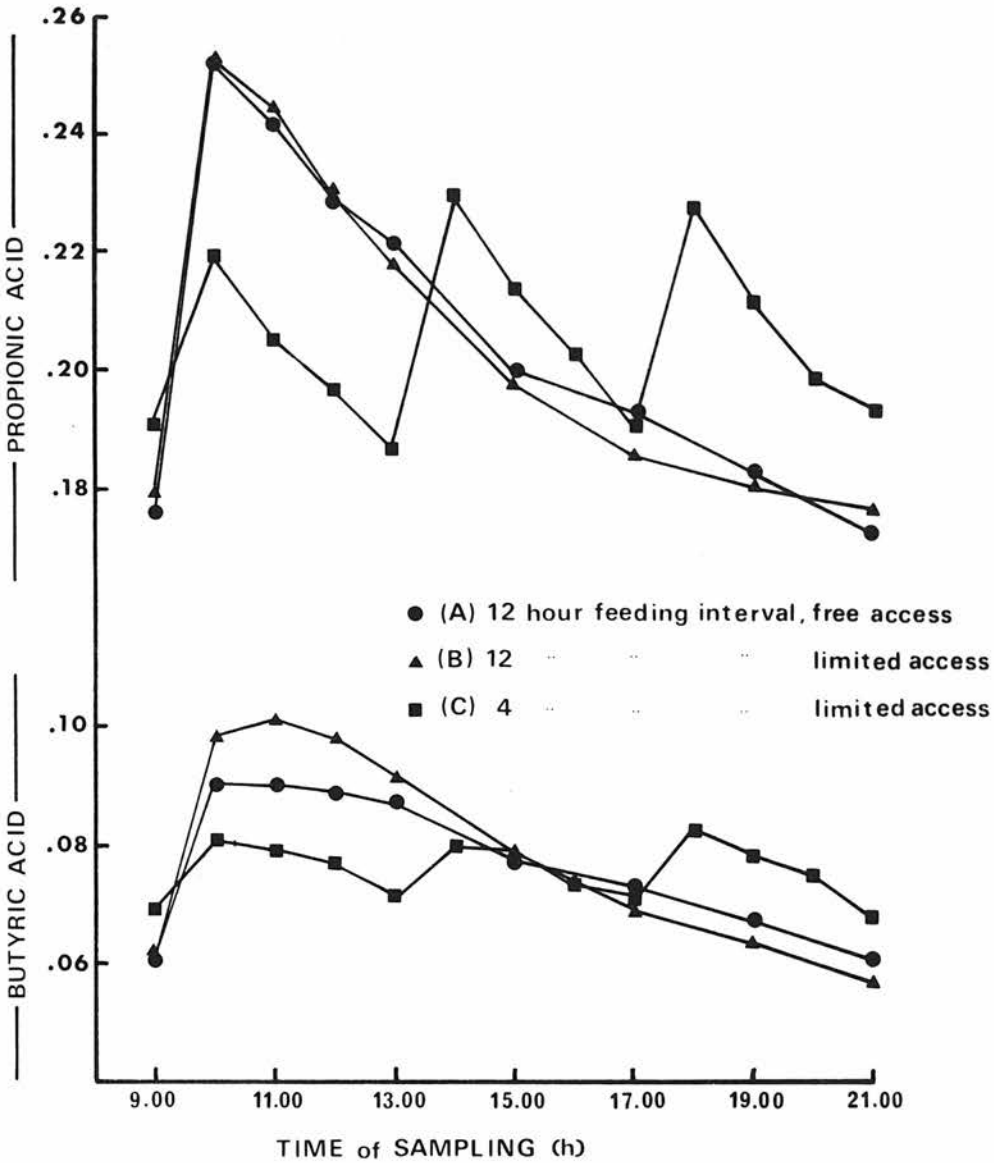

Fig. 25 RUMINAL MOLAR PROPORTIONS OF PROPIONIC AND BUTYRIC ACIDS

86 m mol/l for the four hourly feeding regime is similar to that of Gray et al. (1967) of 64 to 90 m mol/l for two hourly feeding of a chopped alfalfa hay diet. These authors showed a similar condensed curve when the feeding interval was reduced from twelve to two hours. The shape of the twelve hour curve resembled that for the silage when the animals were allowed free access. Satter and Baumgardt (1962), Bath and Rook (1963) and Faichney (1968) all found less fluctuation in rumen TVFA concentration with more frequent feeding, as has been shown in the present investigation with the silage diet.

Major Acids: Fig. 24 shows the molar proportions of acetic acid and Fig. 25 the proportions of propionic and butyric acids in the rumen contents of the sheep on each of the feeding regimes. There was no apparent difference between the minimum and maximum values of propionic acid for the treatments involving twelve-hour feeding intervals. The limited access treatment gave lower minimum values of acetic acid, and a different shape of curve one to two hours after feeding. These are reflected by higher butyric acid values between one and four hours post-feeding. Increasing the frequency of feeding reduces the range of the molar proportions of acetic, propionic and butyric acids in the rumen. Bath and Rook (1965) found the molar proportions of acetic acid were higher when cows were fed indoors than when grazing the same sward, and there were corresponding decreases in butyric acid but little change in propionic acid.

Putnam et al. (1961) found little change in the molar proportions of acetic, propionic and butyric acids with increased frequency of feeding, nor did Faichney (1968) when the feeding frequency was increased from once to eight times daily. Sutherland et al. (1963), although finding increased molar proportions of propionic acid with increase in feeding frequency, could find no significant difference between the two feeding regimes. Hillier et al. (1968), for twice and six times

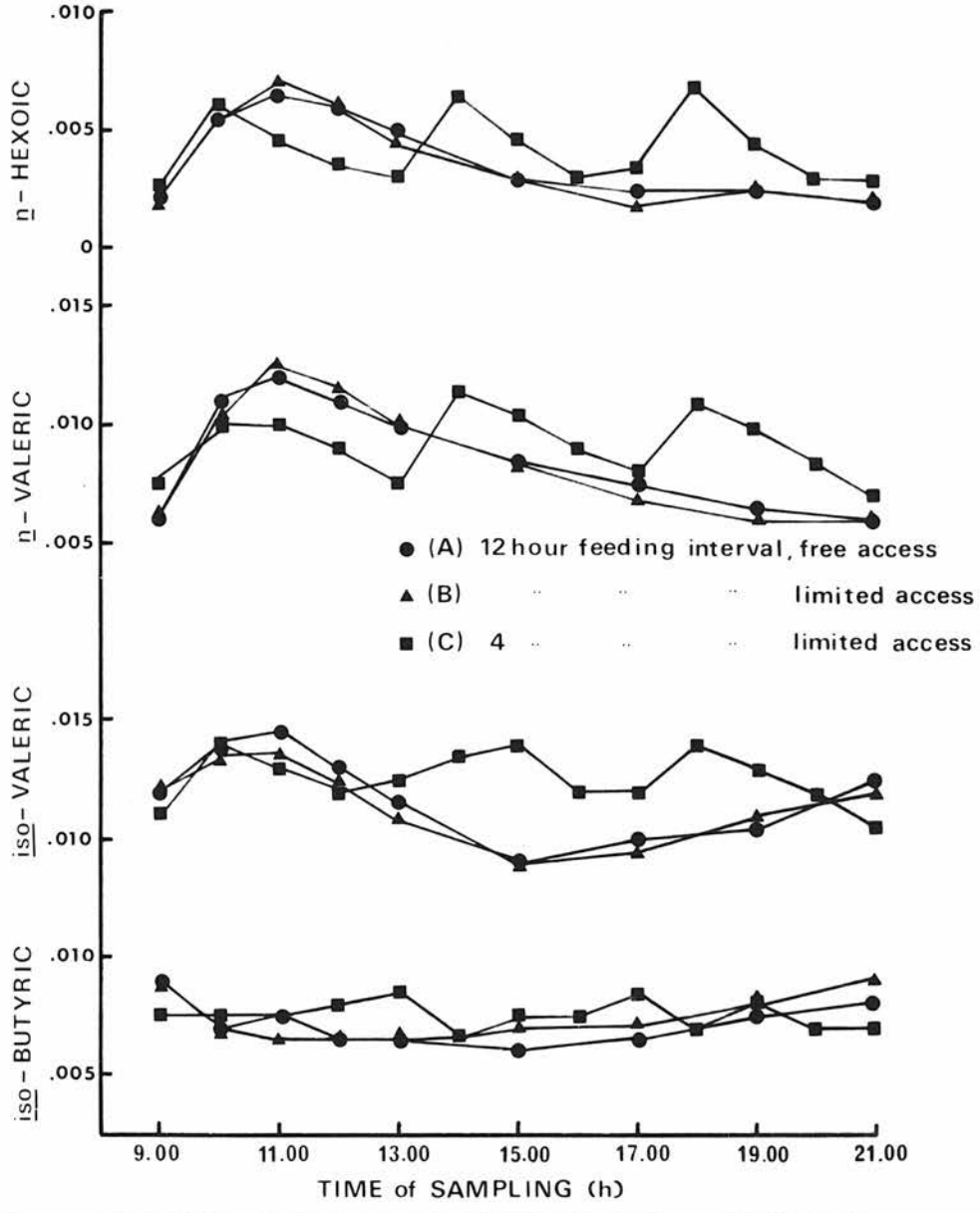

Fig. 26 RUMINAL MOLAR PROPORTIONS OF iso-BUTYRIC, iso-VALERIC, n-VALERIC and n-HEXOIC ACIDS

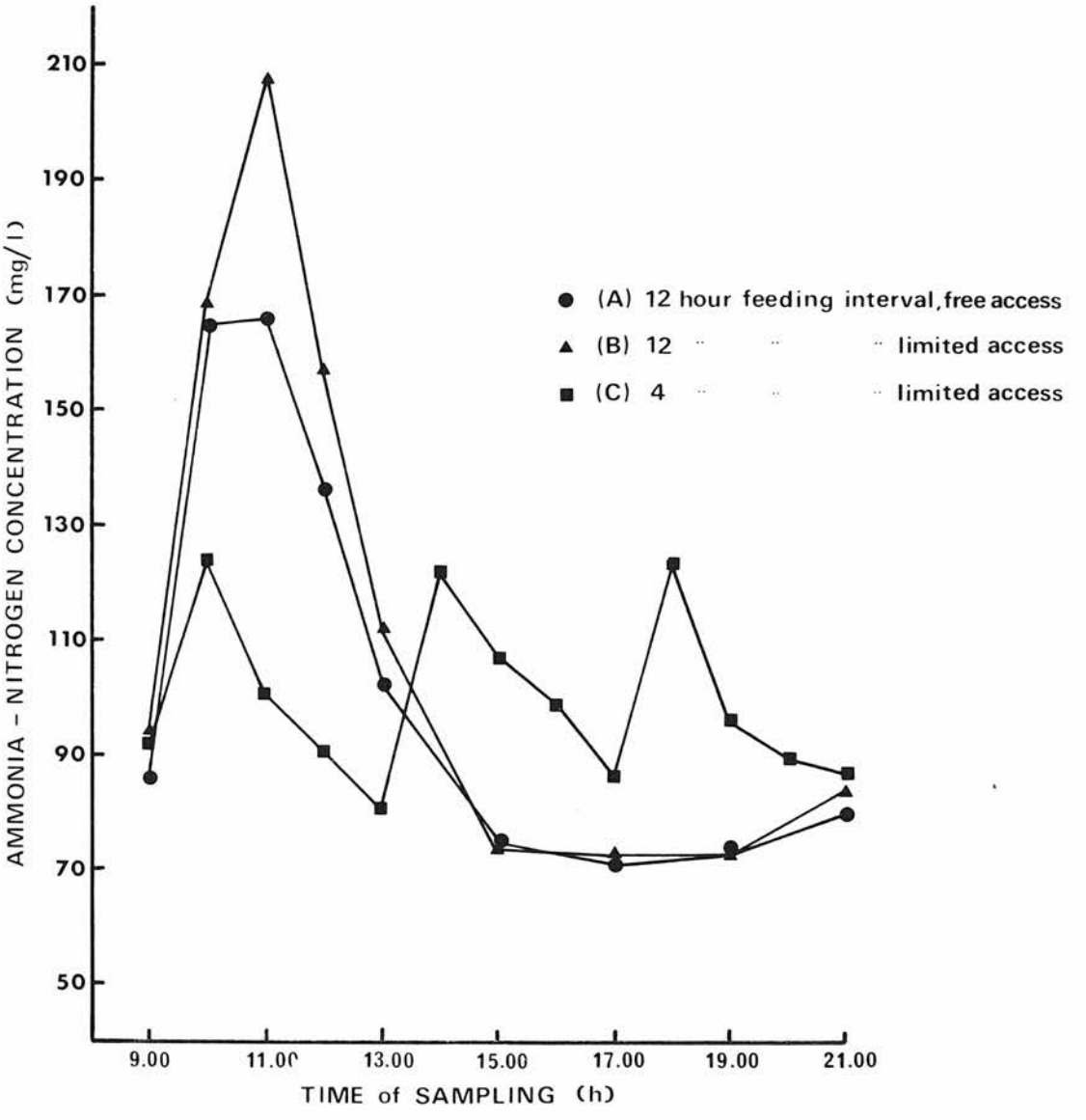

Fig. 27 RUMINAL AMMONIA CONCENTRATION

daily feeding, reported molar proportions of acetic, propionic and butryic acids at four hours post-feeding of 0.643 and 0.663, 0.211 and 0.196 and 0.103 and 0.112 respectively.

Bath and Rook (1963) confirmed that the ranges of proportions of the major volatile fatty acids in the rumen were narrowed by increased frequency of feeding of a hay diet. Knox and Ward (1961), working with a diet of barley and hay, confirmed the lowering of acetate proportions with increased frequency of feeding but did not find a narrowing of the range.

Minor Acids: The curve patterns for the minor ruminal volatile fatty acids (Fig. 26) confirm the effects of the three treatments shown by the major acids, ie. the pattern for the twelve-hour feeding interval is compressed into four hours.

Ammonia: The curves for the concentration of ammonia in the rumen contents are shown in Fig. 27. Minimum pre-feeding values were similar for the three treatments. With twice daily feeding the maximum was higher when access was limited. With frequent feeding maximum concentration was lower than with the other two treatments and occurred at one hour post-feeding.

Moir and Somers (1957) with a diet of concentrate cubes plus chaff reported pre-feeding ruminal ammonia nitrogen concentrations of 66, 77 and 93 and maximum values of 169, 153 and 116 mg/l for once, twice and four times daily feeding respectively. Satter and Baumgardt (1962) compared twice with four and eight times daily feeding, Portugal (1963) twice with continuous, and Sutherland et al. (1963) twice or four times with continuous, and found the fluctuation in ammonia concentration was less when the feeding interval was shorter.

Comparison of Slopes of Concentration Curves for Various Rumen Parameters: Analysis

of variance has been carried out on the effect of the treatments on the first slope of each fermentation curve, that is the change which has taken place between pre-feeding and the first post-feeding sample and on the second slope which is the change occurring between the second post-feeding sample and the subsequent twelve hour pre-feeding sample. The curvatures of the latter were compared using the logarithms of the slopes. The treatment means of the slopes and the significance of the differences are tabulated (Table 13).

Table 13

Summary of Statistical Analysis of Slopes of Rumen Fermentation Curves

Fermentation Characteristic		Treatment Mean A	B	C	A and B	B and C	A and C
pH	1st slope	-0.5	-0.4	-0.2	*	***	**
	2nd slope	-0.04	-0.06	-0.004	**	***	***
	log. 2nd slope	-5.17	-5.15	-4.20	NS	NS	NS
TVFA concentration	1st slope	27.5	25.4	13.6	NS	**	**
	2nd slope	-2.5	-4.0	0.22	**	***	***
	log. 2nd slope	2.93	2.92	4.19	NS	*	*
Acetic acid	1st slope	-11.0	-11.9	-5.1	NS	***	***
	2nd slope	1.00	1.33	0.07	**	***	***
	log. 2nd slope	0.35	0.07	1.03	*	NS	**
Propionic acid	1st slope	8.13	7.11	2.45	NS	***	***
	2nd slope	-0.68	-0.70	-0.05	NS	***	***
	log. 2nd slope	-1.06	-0.08	-0.59	NS	NS	*
Butyric acid	1st slope	2.0	4.2	1.6	NS	NS	NS
	2nd slope	-2.80	-2.13	-1.10	NS	NS	*
	log. 2nd slope	-0.02	-0.01	-0.02	NS	NS	NS

Table 13 (Cont'd)

Fermentation Characteristic		Treatment Mean A	B	C	A and B	B and C	A and C
Ammonia -Nitrogen	1st slope	8.0	7.8	2.7	NS	**	**
	2nd slope	-0.83	-1.05	-0.06	NS	***	***
	log. 2nd slope	1.90	1.90	1.18	NS	**	*

The values marked with asterisks are significantly different:
* $P<0.05$; ** $P<0.01$; *** $P<0.001$; NS = Non significant.

Animal Performance

Kay and Hobson (1963) suggested that ruminants would benefit from more frequent feeding. Mohrman et al. (1959) obtained increased live weight gains with steers fed six times instead of twice daily, but intake was increased by the more frequent feeding. However, Gordon and Tribe (1952), Rakes et al. (1957), Putnam et al. (1961) and Faichney (1968) showed increased live weight gain with increased frequency of feeding when intakes were almost equalised. Rhodes and Woods (1962b), Rakes et al. (1961) and Clark and Keener (1962) did not find any improvement in live weight gain of lambs by increasing frequency of feeding.

Knox and Ward (1961) found the ratio of acetic to propionic acid in the rumen was higher when feeding was twice compared to eight times daily. In the present investigation, with limited access and twice daily feeding the ratios ranged from 4.16 at pre-feeding to 2.44 at minimum acetic acid proportion, while with six times daily the range was from 3.80 to 2.83. Knox and Ward (1961) offer the increased utilization of propionic acid for fattening, quoted by Armstrong et al. (1958), as an explanation for the improved live weight gain. They also suggested this improvement could be due to increased concentration of TVFA. Satter and Baumgardt (1962) indicated that increased frequency of feeding increased the regular

supply of feed materials to the rumen and gave rise to an even production of metabolites. Gray et al. (1967) and Faichney (1968) found no difference in total volatile fatty acid production by increasing the frequency of feeding.

With dairy cows Burt and Dunton (1967) claimed there was no advantage in milk production from feeding more than twice or three times daily. These authors noted the same reaction for cows fed normally as was found with the sheep in the present investigation. When fed twice daily with free access, these animals were stimulated into silage consumption when the extra feeds were given to the more frequently fed sheep in adjacent crates.

CONCLUSIONS

Increased frequency of eating results in condensed ruminal fermentation curves which reduce the likelihood of showing significant differences between treatments, as does the use of mean values to represent the pattern of fermentation.

With twelve hour feeding intervals free access results in erratic eating over the period between meals particularly with individual animals and again reduces the likelihood of showing differences between treatments.

Feeding at twelve hour intervals is, practically, more convenient than at four hour intervals. The increased labour of removing the food residues when access was limited to two hours was justified by the more distinctive fermentation curves obtained under this regime.

The data obtained in this preliminary trial would tend to support the contention by many workers of the practical benefits of increased frequency of feeding. The benefits reflect the improved intake of digestible dry matter and more desirable rumen fermentation patterns to be expected with this technique.

VI

EXPERIMENT 3

THE NUTRITIONAL CHARACTERISTICS OF SILAGES MADE BY DIRECT AND DELAYED SEALING OF THE SILO

INTRODUCTION

Ideally, grass should be ensiled by instantaneous achievement of anaerobiosis within the silo. In practice silage making frequently involves delays in the achievement of air-free conditions. In an attempt to simulate farm conditions, forage harvested ryegrass was piled on the base of two-tonne cylindrical plastic silos with the sides rolled down. The herbage was exposed to the air for seventy-two hours, at the end of which the sides were unrolled over the heap of grass and sealed. A control silage, ensiled into a similar plastic silo but sealed immediately was made from the same grass crop. Dried grass was made from the same source. The fate of these materials when fed to sheep was investigated. The experiment was divided into two sections. 3A used first cut Italian ryegrass of high water soluble carbohydrate and low nitrogen contents. 3B used third cut grass from the same field but with low water soluble carbohydrate and high nitrogen contents resulting from the application of fertilizer two weeks before cutting.

3A.

SILAGES MADE FROM GRASS OF HIGH WATER SOLUBLE CARBOHYDRATE AND LOW NITROGEN CONTENT.

EXPERIMENTAL

The silages and the dried grass were fed to nine sheep in a cross-over design with three sheep on each treatmentduring each period. Sheep were allocated at random to different treatments and the design was balanced for residual effects. The design is shown in Table 14.

Table 14.

Cross-over Design for Three Treatments and Nine Sheep

Sheep No. / Period	1 (414)	2 (434)	3 (409)	4 (449)	5 (448)	6 (437)	7 (435)	8 (447)	9 (680)
I	A	B	C	A	B	C	A	B	C
II	B	C	A	C	A	B	B	C	A
III	C	A	B	B	C	A	C	A	B

The treatments were:-

A. Control silage - 953 g dry matter fed daily to each sheep.

B. Delayed sealed or treated silage - 1060 g dry matter fed daily to each sheep.

C. Dried grass - 880 g dry matter fed daily to each sheep.

The feeding regime was as described earlier. The dry matter content of the diets was 201, 212 and 881 g/kg and the pH values were 4.28, 4.49 and 5.94 for the control silage, treated silage and dried grass respectively. The composition of the materials are shown in Table 15.

Table 15

Composition of Silages and Dried Grass (g/kg dry matter).

	A (Control silage)	B (Treated silage)	C (Dried grass)
Crude protein	113	116	112
Crude fibre	255	255	211
MAD - fibre	316	307	244
Ash	94	86	82
Total nitrogen	18.1	18.5	17.9
Protein nitrogen	5.2	8.5	13.1
Water soluble nitrogen	12.9	10.0	4.8
Volatile nitrogen	2.1	1.4	-
Cellulose	281	271	231
Acetic acid	10	10	-
Propionic acid	4	tr	-
Butyric acid	20	13	-
Lactic acid	88	38	-
Succinic acid	20	16	-
Water soluble carbohydrate	87	140	278
Ethanol	20	16	-

The composition of the silages was similar. The delayed sealing of the silo had resulted in a silage in which the water soluble carbohydrate content was higher, the water soluble nitrogen and volatile nitrogen was lower, the pH was slightly higher and lactic and butyric acids lower than the control silage. The fermentation in the control silage was more extensive.

RESULTS AND DISCUSSION

Nutritive Value

Intake: Mean daily dry matter intakes of the control silage, treated silage and the dried grass were 24.2, 35.4 and 39.9 g/kg $W^{0.75}$ respectively. Owing to the restricted amounts of dried grass provided, the latter figure does not represent the true intake potential of this material. Analysis of variance gave evidence for a significant difference in intake between the three treatments. Mean daily intakes for individual sheep on each treatment are given in Appendix Table 42.

Harris and Raymond (1963) reported results which differed from the present investigation when they fed two wilted ryegrass silages, one ensiled after a forty-eight hour delay and the other ensiled directly. Intakes when fed to sheep were 56.3, 54.2 and 56.5 g/kg $W^{0.73}$ for the control and treated silages and barn dried hay respectively. The dry matter of the silages was 299 and 306 g/kg respectively. However, in a similar experiment with unwilted grass, Harris et al. (1966) reported an increase in intake with delayed sealing of the silo. They quoted mean daily intakes for sheep of 46.9 and 56.0 g/kg $W^{0.73}$ for directly ensiled and delayed sealed silage respectively.

Waldo et al. (1966) reported reduced intakes when heifers were fed silage compared with hay on an ad libitum system. Moore et al. (1960), Gordon et al. (1961) and Strickland et al. (1966) all reported reduced intakes of silage compared to hay. The reduced intake of silage has been attributed to its moisture content (Dodsworth, 1954), but the data of Mahapatro and Leffel (1964) and Baile and Mayer (1970) do not support this view, that water content per se reduces dry matter intake. Harris and Raymond (1963) showed a reduction in intake when silage replaced fresh grass diets.

Many workers, (Montgomery et al., 1963; Simkins et al., 1965; Ulyatt, 1965; Baile and Pfander, 1966; Weston, 1966; Bhattacharya and Warner, 1968), have reported decreased intake of feed when volatile fatty acids were injected into the rumen of cattle and sheep. However, Senel and Owen (1966) reported increased dry matter intake when lactate and acetate were added to the basal hay diet to simulate highly fermented silages. McLeod et al. (1970) reported a decrease in intake when the free acid content of silage was increased by the addition of lactic acid, but an increased intake when the free acid content was neutralised.

The lower intake of the control compared with the treated silage in the present work probably reflects the more extensive fermentation in the control silage with subsequent lowering of water soluble carbohydrate, a higher content of fermentation acids, and a greater content of water soluble and volatile nitrogen.

Digestibility: McDonald et al. (1960) suggested that the lower intakes, by sheep, of silage compared with fresh grass was unlikely to affect the digestibility of the individual constituents of the food. Jackson and Anderson (1968) showed little effect on metabolisable energy values of wilted and unwilted silages when fed at three levels of intake. In the present investigation statistical analysis of the digestibility data to compare the treatments involved analysis of covariance and showed that in this experiment the dry matter intake had little effect on the mean treatment values for digestibility of dry matter, organic matter and nitrogen, or metabolisable energy.

Mean values for the digestibility data for nine sheep on each treatment are given in Table 16. The values for the individual sheep are given in Appendix Table 42.

Table 16

Nutritive Value of the Silages and Dried Grass

	A (Control silage)	B (Treated silage)	C (Dried grass)
Digestibility of dry matter	0.776	0.733	0.752
Digestibility of organic matter	0.808	0.770	0.779
Digestibility of nitrogen	0.719	0.636	0.639
Metabolisable energy (MJ/kg)	13.4	12.6	10.1
Gross energy of digestible organic matter (MJ/kg)	22.7	21.6	17.4

Analysis of covariance did not give significant evidence for any differences in digestibility of dry matter or organic matter between the treatments. Differences between the control silage and the other two treatments in the digestibility of nitrogen were significant ($P < 0.05$). The lower values for the treated silage and the dried grass compared with the control silage were perhaps due to the heating involved in their production. There was a significant difference between the metabolisable energy values.

The absence of a significant difference in digestibility of dry matter in the present investigation is in agreement with the findings of Harris and Raymond (1963) who found no difference in digestibility of dry matter between silages made by direct and delayed sealing, or with barn dried hay made from the same sward. Campling (1966) also found similar digestibilities of hay and silage. McDonald et al. (1966) found no difference in digestibilities of nitrogen or organic matter between silages made at different fermentation temperatures, nor did they find (1968) differences between silages made from the same crop when the composition

of the dry matter was different. McDonald et al. (1962) with Italian ryegrass found no differences in digestibility of organic matter and nitrogen between grass and directly made silage, but with cocksfoot reported that digestibility of nitrogen was higher for silage than the grass. Thomson (1968) found a lower digestibility of nitrogen for silage compared with hay or grass.

The higher digestibility of the control silage in the present work may be due to the higher content of simpler compounds resulting from the more extensive fermentation taking place in this product.

The higher gross energy of the digestible organic matter which was found in this experiment is in keeping with the observations of Alderman et al. (1971) and McDonald et al. (1973). The latter point out that fermentation leads to an increase in high energy reduction compounds such as ethanol and mannitol. This explains the higher gross energy of the control compared with the treated silage. The higher metabolisable energy of the control is explicable in terms of its higher energy of digestible organic matter and its increased digestibility.

Rumen Characteristics

The data on rumen fermentation characteristics have also been adjusted for level of intake effect by analysis of covariance with dry matter intake g/day as the covariate. The curves illustrating the changes in rumen fermentation characteristics with time of sampling after feeding for each treatment are based on adjusted mean values for nine sheep. A statistical comparison has also been made of these treatment curves on the basis of first slope considered to be from the pre-feeding to the one hour post-feeding sample, the maximum of parabola for time of attainment and value, and the second slope considered to be from three hours post-feeding to the next pre-feeding sample.

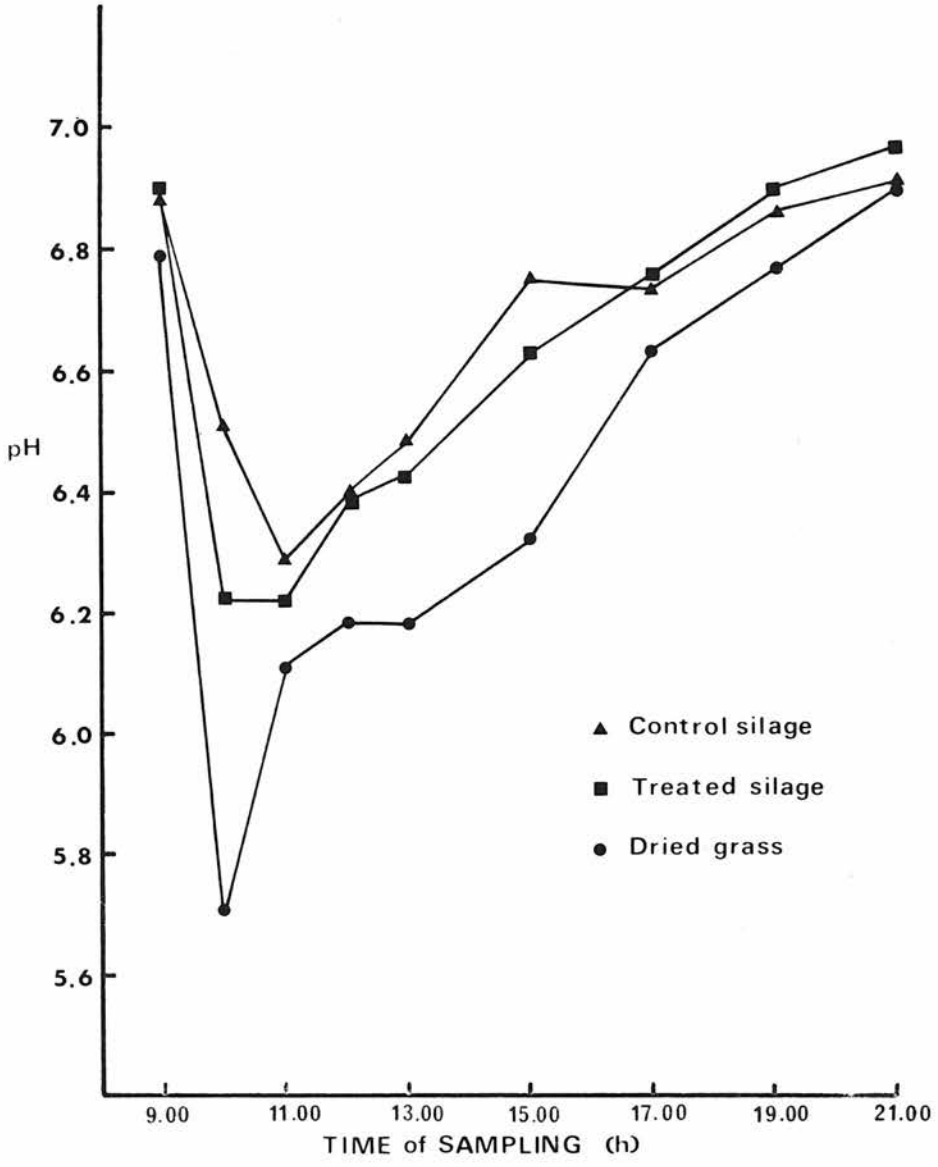

Fig. 28 RUMINAL pH VALUES

The composition of rumen contents for the individual sheep on each diet is shown in Appendix Tables 43-69.

pH and TVFA Concentration: Fig. 28 shows the changes in pH of the rumen contents of the sheep for the three treatments. Analysis of covariance showed that the differences between the dried grass and the silages at 10.00 h. were significant as was that between the grass and the control silage at 15.00 h. There was no significant evidence for differences in ruminal pH values at any other sampling stage. Differences in the first slope between the three diets were significant. Differences between the control silage and the dried grass in the minimum of parabola for concentration, and the second slope, were also significant.

The similarity of the curves for the silage treatments reflects the similar nature of dry matter of these diets. However, it might have been expected that the higher water soluble carbohydrate content of the treated silage would have caused a higger difference in minimum pH values of the rumen contents than was actually found. The higher minimum ruminal pH values with the silage diets compared with the dried grass may reflect their higher buffering capacities of 900 and 990 for the control and treated silages compared with 265 (m equivalent /kg dry matter) for the dried grass, as well as their lower content of water soluble carbohydrate.

The adjustment in rumen pH for the level of intake effect by analysis of covariance necessitated lowering the pH for the low intake of the control silage and raising that for the dried grass with the highest intake. This is in keeping with the results of Bath and Rook (1963), Terry and Tilley (1963), Fenner et al. (1967) and Rumsey et al. (1970) who found increased intake of all roughage diets lowered the pH of rumen contents.

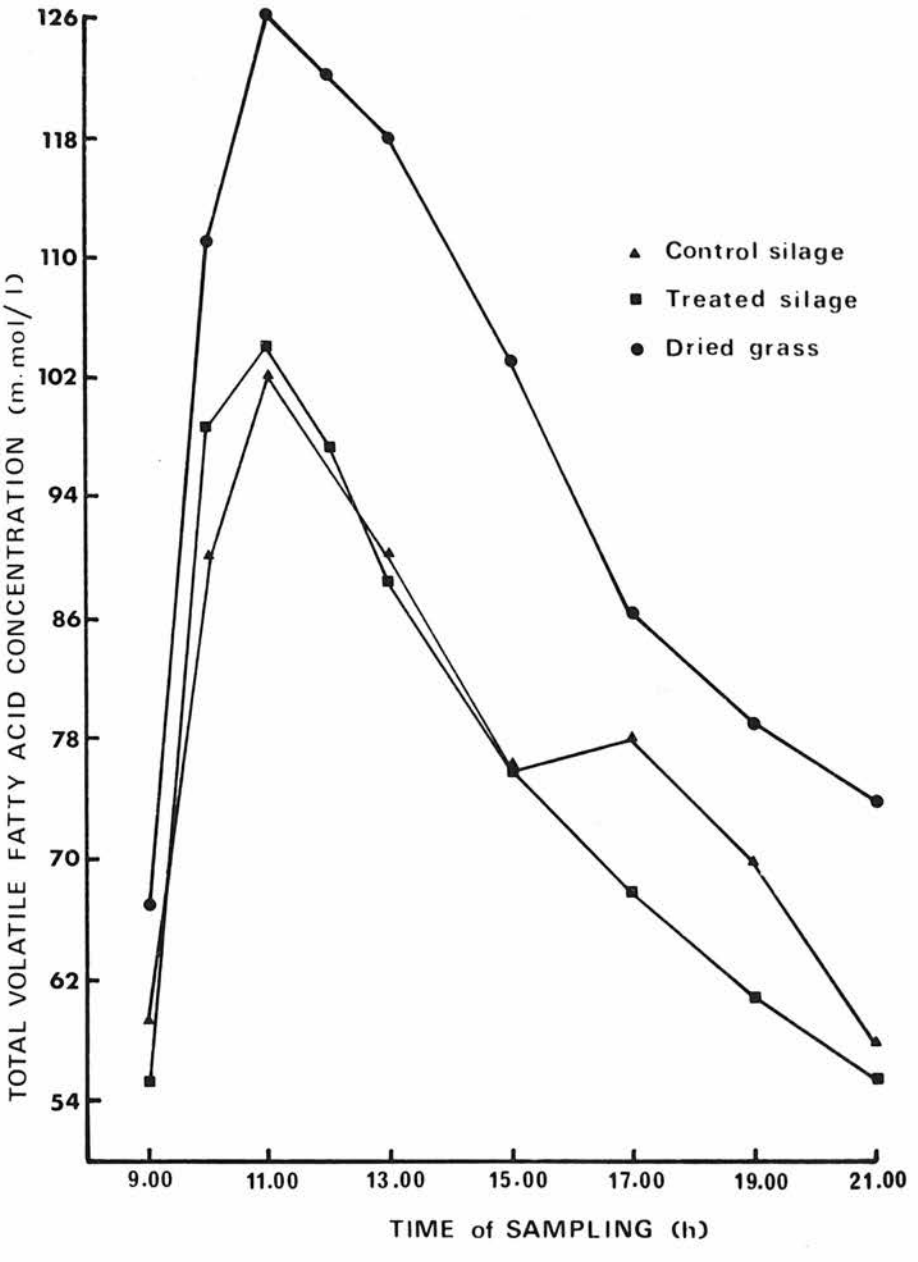

Fig. 29 RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATIONS

Christian and Williams (1957) for a dried grass diet reported a narrower range of rumen pH values, from 6.8 to 6.6, than those found in the presenttrial. Using mean values from several rumen samples Bath and Rook (1965) and Anderson and Jackson (1971) compared hay with silage made from the same grass sward. The former workers quoted pH values of 6.01 and 6.21, and the latter 6.34 and 6.10 for silage and hay respectively. Fenner et al. (1970) compared hay with maize silage and found little change in average rumen pH values. The results for the silage treatments in the present investigation are in agreement with those of Schambye and Phillipson (1949) who quoted a range of 6.7 to 6.2 for a hay plus meal diet. They are also similar to those for an arable silage diet of 6.25 and 6.90 quoted by Balch and Rowland (1957).

The pattern of post-feeding change of ruminal pH in the present investigation is similar to that shown by Steger et al. (1970) for a hay diet. They gave a maximum value of 7.1 before feeding and a minimum value two to four hours post-feeding of 6.2. For a grass diet, the pre-feeding value was 6.7 and the minimum value, which occurred at three hours post-feeding, was 5.9.

Ruminal pH values in the present investigation were generally lower for the dried grass than either silage, but the values for the treated silage were only marginally lower than those for the control. The size of the initial drop in pH (0.38, 0.68 and 0.99) following ingestion of food appears to be related to the water soluble carbohydrate content of the diet (87, 140 and 278 g/kg).

Fig. 29 illustrates the changes in TVFA concentration with time of rumen sampling for the three treatments. Analysis of covariance showed that differences in TVFA concentration between the dried grass and both silage treatments at 13.00, 15.00 and 21.00 h. and between the treated silage and grass at 17.00 h. were significant. There was no significant evidence for differences between the treatments in the comparison of slopes, maximum of parabola or at other sampling stages.

Minimum ruminal TVFA concentrations for all treatments were at pre-feeding. Maximum concentrations occurred about two and a half hours post-feeding. Concentrations were higher for the dried grass diet at all sampling times. With all treatments the TVFA concentration curves reflect those of pH, the food with the highest sugar content producing the highest rumen TVFA concentration.

The corrections applied to the TVFA concentrations to allow for level of intake effect by the covariance analysis of the present data are in keeping with the results of Bath and Rook (1963), Terry and Tilley (1963), Fenner et al. (1967) and Rumsey et al. (1970) who reported increased TVFA concentration with increased intake of all roughage diets.

Christian and Williams (1957) fed a dried grass diet and reported a range of ruminal TVFA concentration of 65 to 80 m mol/l compared with 67 to 127 in the present investigation. This is similar to that of 70 to 124 m mol/l quoted by Schambye and Phillipson (1949) for a hay plus meal diet. El Shazly (1952) quoted 96 to 142 m mol/l for a dried grass and 65 to 128 m mol/l for a silage diet. Balch and Rowland (1957) reported 87 to 130 m mol/l for an arable silage diet. These are rather higher than the range of 55 to 104 m mol/l for silage diets in the present investigation.

Bath and Rook (1965) showed the average ruminal TVFA concentration was higher for silage compared with a hay diet, while Anderson and Jackson (1971) found slightly lower values for silage. Fenner et al. (1970) found little change in TVFA concentration when comparing hay with silage, as did Anderson and Jackson (1971) with grass and silage. Williams and Christian (1959) with four different silages found maximum TVFA concentration at one hour post-feeding for all the diets and a narrower range of values than in the present work. Minimum values ranged from 35 to 41 and maximum from 49 to 62 m mol/l but dry matter intakes of the silages were low from 137 to 151 g/feed.

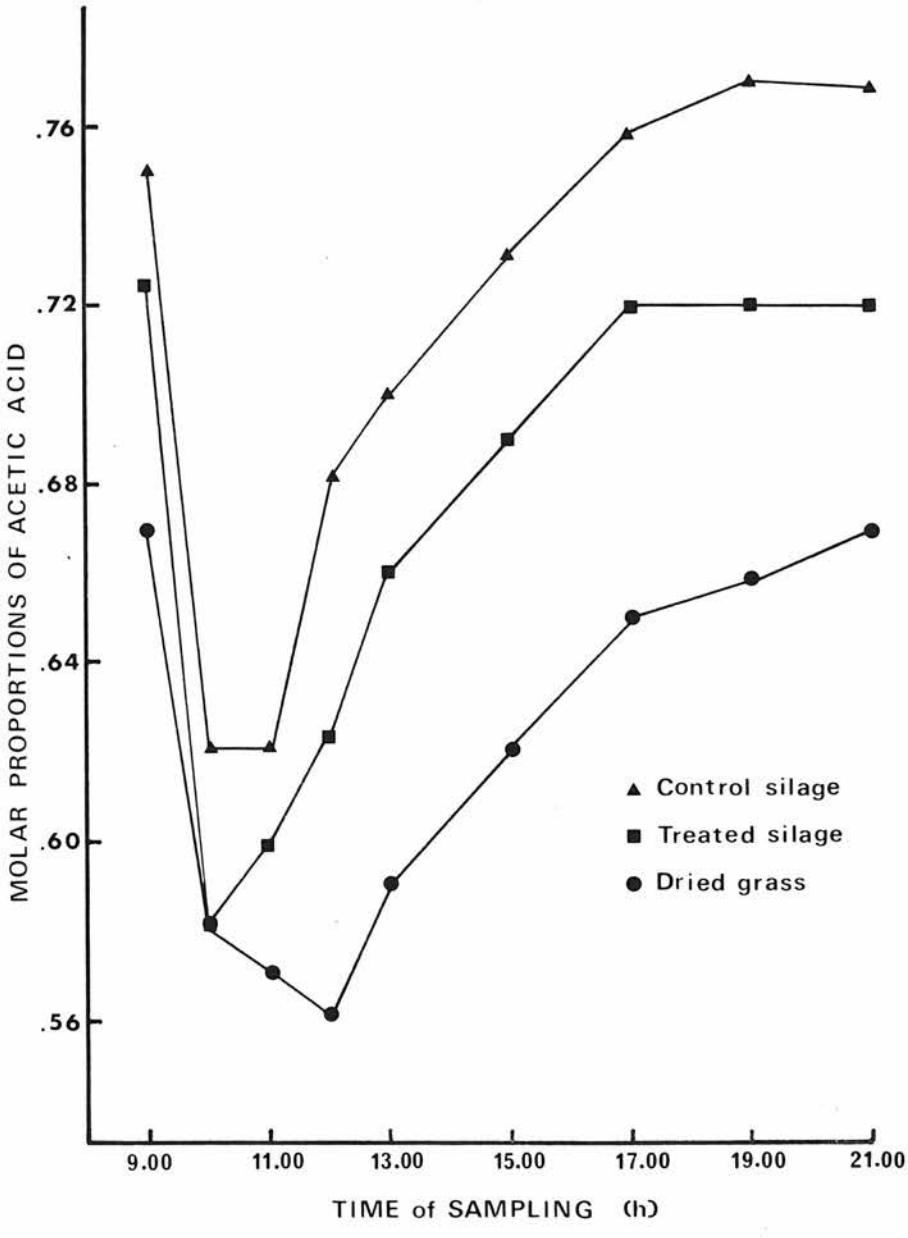

Fig. 30 RUMINAL MOLAR PROPORTIONS OF ACETIC ACID

Major Acids: Terry and Tilley (1963) found that the molar proportions of acetic acid in the rumen decreased when the intake of an all roughage diet was increased. Bath and Rook (1963) with a hay plus concentrate diet confirmed this and found the proportions of propionic and butyric acid were correspondingly increased. Rumsey et al. (1970) found less change in the ratio of acetic to propionic acid when intake was increased, if the diet was a concentrate compared to a roughage. Fenner et al. (1967) fed hay at four levels of intake and could find no relationship between intake and the molar proportions of acetic, propionic and butyric acids in the rumen. Bath and Rook (1963) showed that by increasing the intake of hay from 22 to 33 kg the molar proportions of acetic acid in the rumen increased from 0.678 to 0.705 and the propionic decreased from 0.190 to 0.178. This agrees with the results of the covariance analysis in the present trial, since the low level of intake was adjusted by increasing the molar proportion of acetic acid and decreasing the molar proportion of propionic acid in rumen contents.

Fig. 30 shows the molar proportions of acetic acid and Fig. 31 of propionic and butyric acids in the rumen contents of sheep fed the silages and the dried grass.

The proportions of acetic acid were lowest for the dried grass and highest for the control silage throughout the sampling cycle. Maximum values were at pre-feeding for all treatments. There was no significant evidence for differences between treatments in their minimum values or of the time of achievement of minimum values. The times of minima of parabola were 2.11, 2.19 and 2.54 h., with values of 0.630, 0.588 and 0.557 for the control and treated silages and dried grass respectively.

Analysis of covariance of the molar proportions of acetic acid in the rumen samples showed the differences between the dried grass and the control silage at 09.00, 12.00, 13.00, 15.00, 17.00, 19.00 and 21.00 h. were significant. Differences between the treated silage and the other two treatments at 12.00 h. and between the

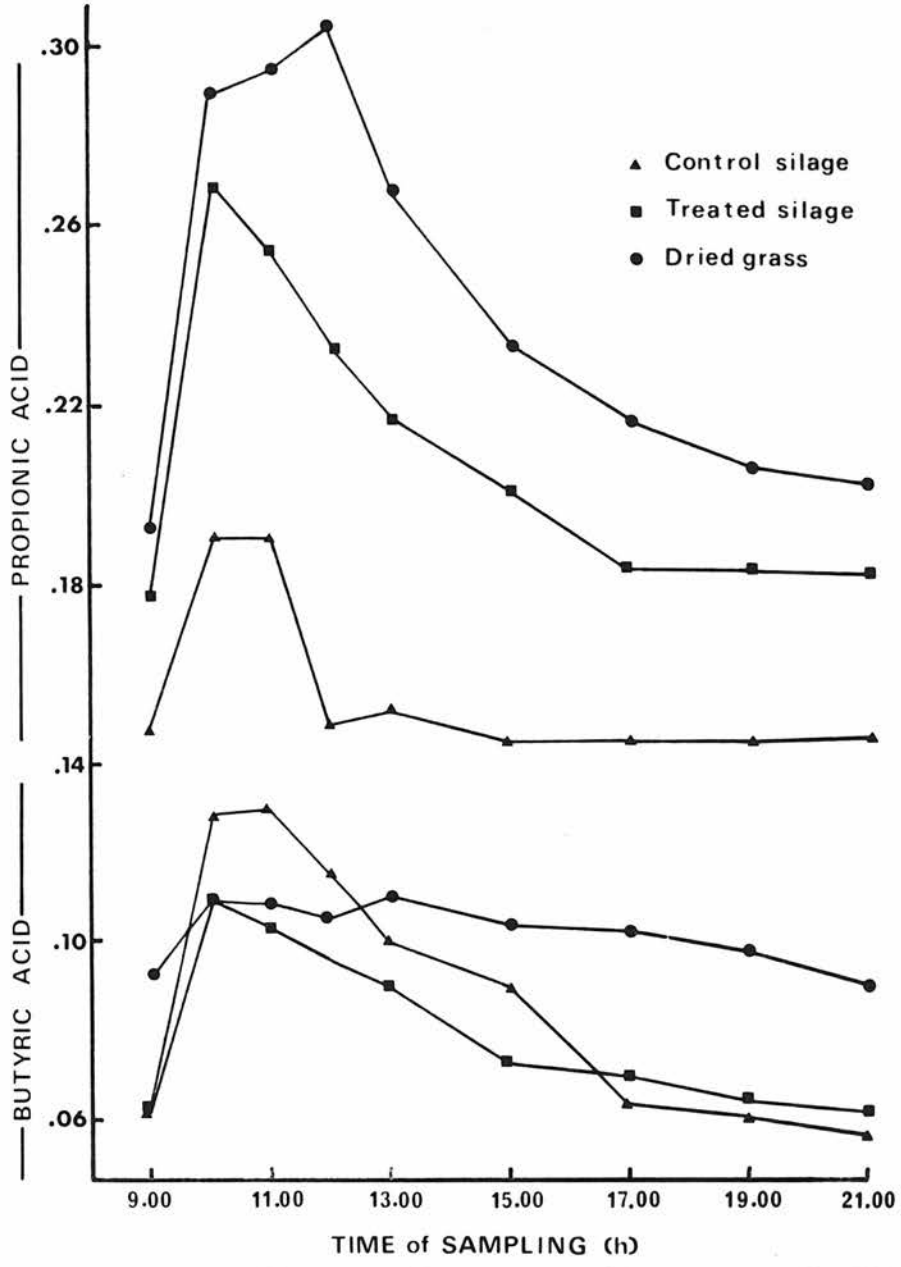

Fig. 31 RUMINAL MOLAR PROPORTIONS OF PROPIONIC AND BUTYRIC ACIDS

treated silage and the grass at 09.00, 13.00 and 17.00 h. were significant.

Differences between the molar proportions of ruminal propionic acid for the three diets were significant at 12.00 h. The molar proportions of propionic acid in the rumen contents were highest with the grass diet and lowest with the control silage. Differences between the control silage and the other two treatments in the maximum of parabola for concentration, and between the dried grass and the other two treatments in the time of attainment of maximum concentration were significant. Differences between the dried grass and the other two treatments in the second slope of the curves were also significant.

Analysis of variance of the molar proportions of butyric acid in the rumen contents showed the differences between the dried grass and both silages at 09.00, 17.00, 19.00 and 21.00 h. were significant. There was no statistical evidence for differences between treatments in the maximum of parabola or in the first or second slopes of the curves. Minimum values were at pre-feeding and were lower for the silages than the grass.

In the present investigation the molar proportions of acetic acid in the rumen contents of the sheep fed the treated silage are similar to the values for silage quoted by Bath and Rook (1965) of 0.626 and by Anderson and Jackson (1971) of 0.616, but those for the control silage diet are higher. The pre-feeding value of ruminal acetic acid for the treated silage was similar to that for a silage diet quoted by El Shazly (1952) but his pre- and post-feeding range of values was narrower than for either of the silage diets in the present work. The patterns of change in acetic acid following the feeding of the three diets are similar to those of Puech et al. (1968) for silage diets, although their changes are less extensive. Thus, in the present investigation, the ranges were from 0.75 to 0.63 for the control, and 0.73 to 0.59 for the treated silage, compared with 0.72 to 0.63 given by Puech et al. (1968), for unwilted lucerne. The values for the dried grass diet in the

present investigation are similar to those quoted by Anderson and Jackson (1971) for first cut hay, and the range is similar to that found by El Shazly (1952) for a dried grass diet.

The molar proportions of propionic acid in the rumen contents of the sheep fed the control silage in the present investigation are similar to those for silage diets reported by Bath and Rook (1965), while those for the treated silage are similar to those of Anderson and Jackson (1971). The pre-feeding ruminal propionic acid value quoted by El Shazly (1952) for a silage diet is similar to those for the control silage in the present trial, while his maximum value of 0.24 although higher than this control (0.19), is not as high as that for the treated (0.27). The ruminal propionic acid proportions for the dried grass diet in the present work are similar to those for first cut hay diets given by Anderson and Jackson (1971). The post-feeding values for the dried grass diet quoted by El Shazly (1952) were lower than those for the dried grass in the present investigation. It is interesting to note that the range of pre- and post-feeding values of ruminal propionic acid for a dried grass diet quoted by El Shazly (1952) was narrower than that for silage diets, while the opposite effect was found for the control silage and the dried grass, in the present work. The patterns of change in the molar proportions of propionic acid in the rumen contents when the three diets were fed were similar to those reported by Puech et al. (1968), for a silage diet. They reported a minimum pre-feeding value of 0.17 and a two hour post-feeding maximum of 0.22. In the present work the range for the control silage was from 0.15 to 0.19 and for the treated silage from 0.18 to 0.27.

The molar proportions of ruminal butyric acid for the silage diets in the present investigation were similar to those for silage diets reported by Anderson and Jackson (1971) but lower than those of Bath and Rook (1965). The ranges of post- and pre-feeding values were wider than those of El Shazly (1952). Puech

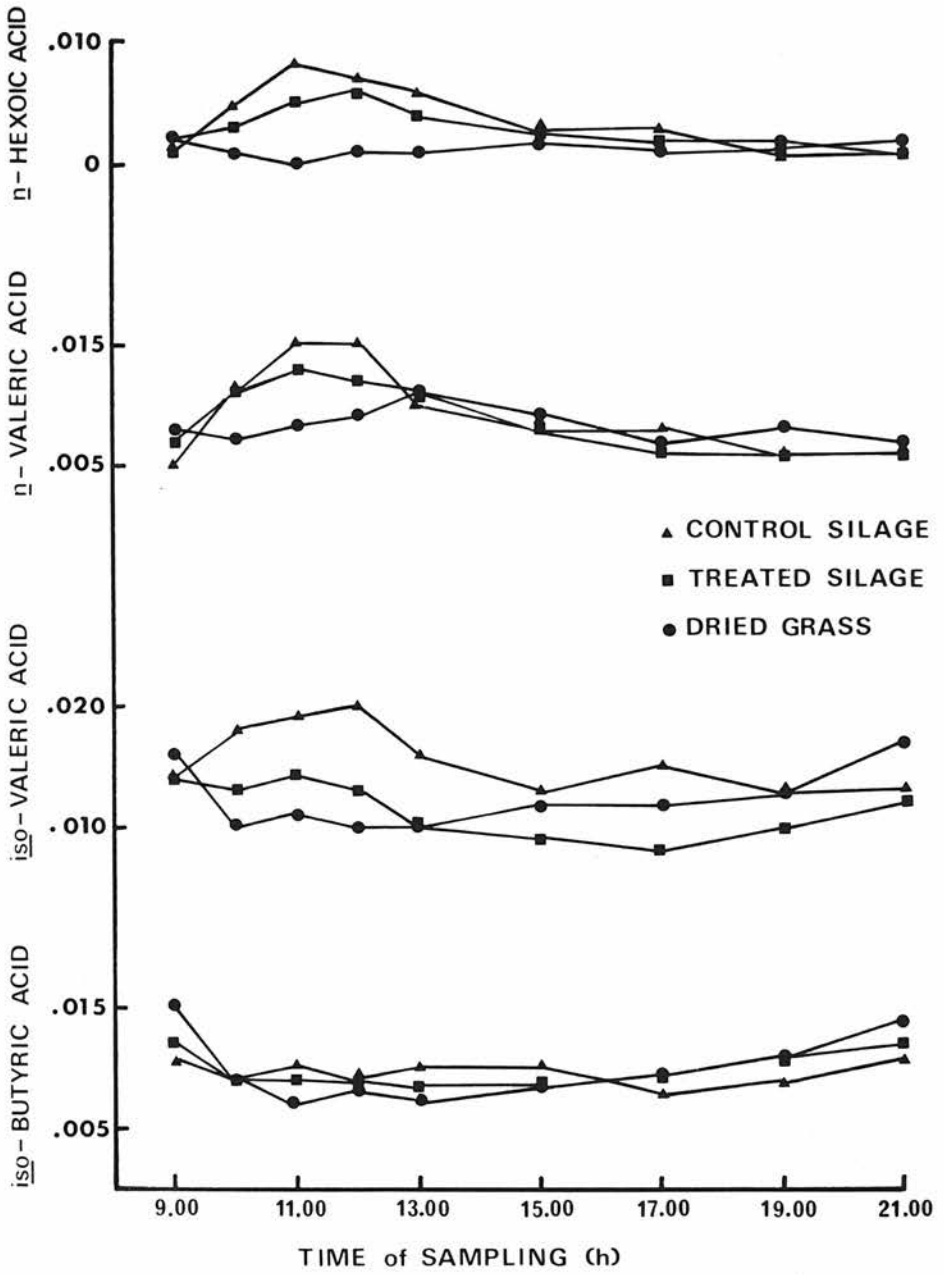

Fig. 32 RUMINAL MOLAR PROPORTIONS OF iso-BUTYRIC, iso-VALERIC, n-VALERIC and n-HEXOIC ACIDS

et al. (1968) reported a pre-feeding value of 0.085 and a two hour post-feeding maximum of 0.125. The corresponding values for the control silage in the present work was 0.07 and 0.13 and, for the treated silage, 0.06 and 0.11. The values of ruminal butyric acid for the dried grass diet in the present work were similar to those of Anderson and Jackson (1971) for a hay diet, and to those of El Shazly (1952) for a dried grass diet.

The rumen fermentations obtained with the dried grass and treated silage diets were more propionic acid oriented than those obtained with the control silage, and this reflects their water soluble carbohydrate and MAD-fibre contents. The proportions of acetic and propionic acids in the rumen contents at periods of maximum fermentation activity, and the corresponding high propionic acid levels, are such as would lend themselves to high efficiency for growth and fattening, but only in the case of the control silage do they approach those required for maximum efficiency ofutilization for milk production.

It is interesting that in the case of the dried grass the fall in the proportion of acetic acid in the rumen acids following feeding was compensated by a rise in propionic acid, while with silages changes in acetic acid proportions were balanced by changes in the sum of the concentrations of propionic and butyric acids.

Minor Acids: Fig. 32 shows the molar proportions of iso-butyric, n- and iso-valeric and n-hexoic acids in the rumen contents of the sheep on the three treatments. Analysis of variance gave no significant evidence for differences between the treatments in the proportions of iso-butyric and iso-valeric acids at any sampling stage, in the first or second slopes of the curves or in the values or time of maximum of parabola. Statistical comparisons of the molar proportions of n-valeric acid in the rumen contents showed significant evidence for differences between treatments at 12.00 h. and between the dried grass and the silages at 11.00

h., but these were small and nutritionally not important. Differences between the molar proportions of n-hexoic acid for the dried grass and the other two treatments were significant at 11.00, 12.00 and 13.00 h.

Fenner et al. (1970) found that there was no difference in the molar proportions of iso-butyric acid in the rumen contents when silage replaced hay, which is in agreement with the present findings when silage replaced dried grass. In this case maximum values of ruminal iso-butyric acid for all diets were at pre-feeding and these declined to a plateau between one and eight hours post-feeding which is similar to the pattern shown by Schmekel (1967) for grass silage diets. The latter worker found maximum values for ruminal iso-valeric acid at the pre-feeding stage and minimum values at six to nine hours post-feeding, which is similar to that for the treated silage in the present investigation. Fenner et al. (1970) reported maximum concentration at three hours post-feeding for maize silage which is similar to the time of maximum for the control silage.

Schmekel (1967), for grass silage diets, reported minimum molar proportions of n-valeric acid in the rumen contents at six hours or at pre-feeding and maximum values at three hours post-feeding. In the present investigation peak values were at two hours post-feeding for both silages but with the dried grass there was a different pattern with a maximum at five hours post-feeding.

El Shazly (1952) compared the molar proportions of n-valeric acid, iso-valeric acid and iso-butyric acid in rumen contents with dried grass and silage diets and reported higher pre- and post-feeding values for the silage diets. Bath and Rook (1965) also found the average values for these acids were higher with a silage diet compared to hay. The results of Anderson and Jackson (1971) were not so clearly defined. The present study confirms the views of the first two groups of workers.

The higher minor acids of the rumen (iso-valeric, n-valeric and n-hexoic) contributed a greater proportion of the total rumen acids with the control silage

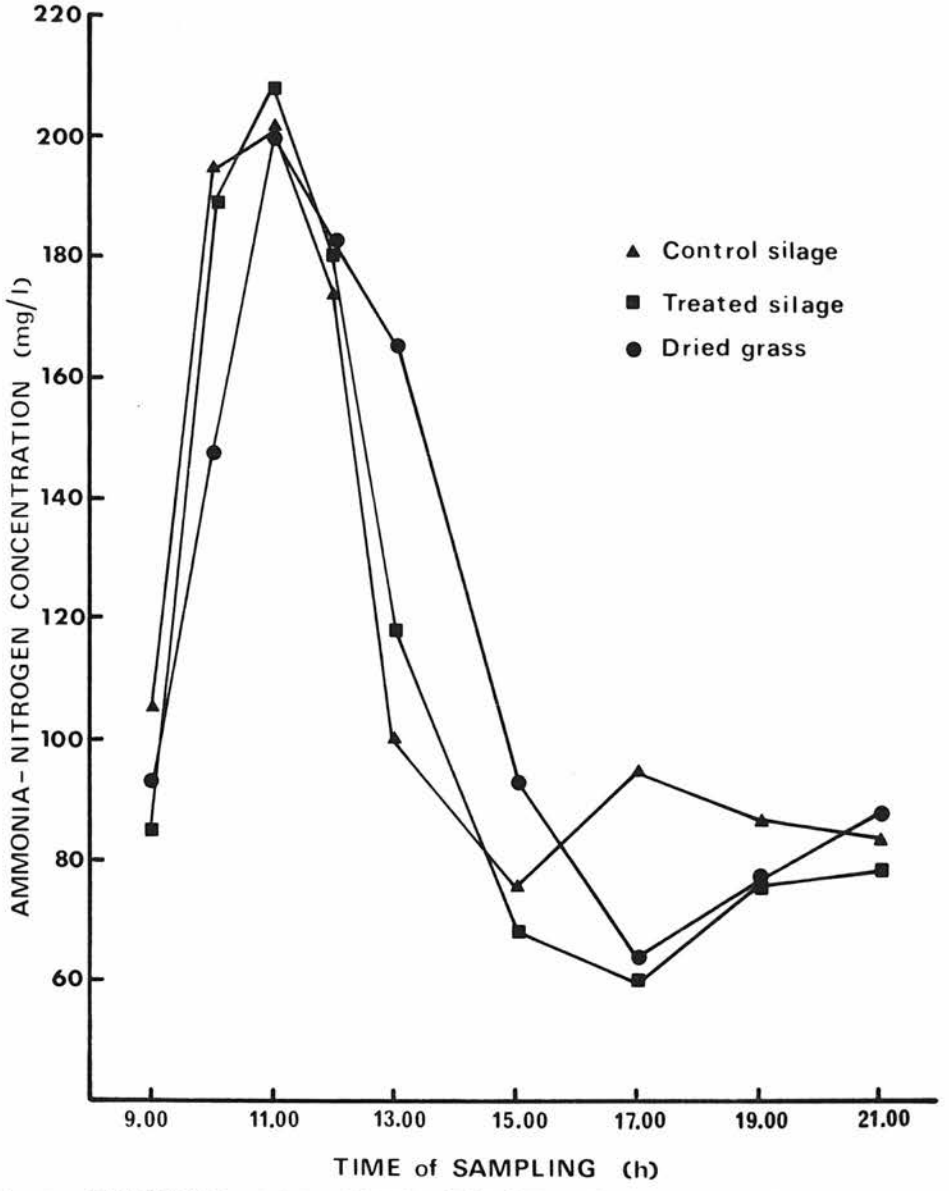

Fig. 33 RUMINAL AMMONIA CONCENTRATION

diet than with the other two diets, probably reflecting the effect on the composition of the more extensive fermentation of the grass during ensilage with the former diet.

Ammonia Concentration: Fig. 33 shows the concentration of ammonia in the rumen contents of the sheep fed the three diets. Analysis of variance gave no significant evidence for differences between the three treatments according to any of the criteria used.

The range of ruminal ammonia nitrogen concentrations for the three diets in the present work, 60 to 208 mg/l, is similar to those reported by Somers (1961), of 46 to 249 mg/l, when a diet of chaff, cubes and meal was fed. The similarity in maximum concentration for the dried grass and the silages is not in agreement with the views of Chalmers (1963) who reported higher concentrations for silage which she suggested were due to:-

a) the formation of soluble nitrogenous compounds from the grass protein during ensilage,

b) a loss of water soluble carbohydrates to acids which were less readily available to the rumen microorganisms and,

c) the denaturing, even by low temperature heating, of the proteins of grass during the drying.

Sutton and Vetter (1971) on the other hand reported a slightly higher minimum and maximum ruminal ammonia concentration when hay was compared with silage.

Williams and Christian (1959) reported maximum ruminal ammonia nitrogen concentrations of between 240 and 170 mg/l at one hour post-feeding for four silage diets. Their highest value, compared with 200 and 208 mg/l in the present study, was from the silage with the highest nitrogen content (28 g/kg).

The pattern of change in ruminal ammonia concentration, following ingestion

of food in the present investigation, is similar to that reported by Durand et al. (1968). For directly ensiled lucerne silage they found a two hour post-feeding maximum ammonia nitrogen concentration of 420 mg/l and a minimum pre-feeding value of 120 mg/l. Their higher range of values reflects the crude protein content of 184 g/kg for the lucerne silage compared with 113 g/kg in the present work. The relatively low ruminal ammonia values obtained here probably also reflect the comparatively limited breakdown of the nitrogen fraction during ensilage. It may have been due to this, that the differences, which might have been expected according to Chalmers (1963), were not obtained. It is difficult, however, to explain why the higher water soluble carbohydrate of the dried grass apparently failed to stimulate microbial activity and lower ruminal ammonia concentration.

Blood Characteristics

Blood pH, plasma glucose and plasma urea values for the nine individual sheep are given in Appendix Table 70.

pH: Blood pH values were 7.33, 7.35 and 7.33 for the control silage, treated silage and dried grass respectively. Analysis of variance gave no significant evidence for differences between the treatments. L'Estrange and Murphy (1972) reported reduced blood pH values from 7.47 to 7.39 in sheep after eighteen days on a diet of grass meal to which mineral acids had been added. Ruminal pH values were reduced by as little as 0.2 units. Although in the present experiment ruminal pH values for the dried grass at minimum values were 0.52 units lower than the treated silage, the blood pH values did not vary between the diets.

Glucose: Plasma glucose concentrations were 542, 555 and 554 mg/l for the control

silage, treated silage and dried grass respectively. Analysis of variance gave no significant evidence for differences between the treatments. Reid (1968) quoted pre-feeding plasma glucose levels in fed non-pregnant sheep of 550 to 650 mg/l. Neither White et al. (1957) nor Williams and Christian (1959b) found differences in blood sugar concentration when the intake of dried grass, by sheep, was reduced. Ross and Kitts (1973) found no difference in plasma glucose concentration in sheep fed hay, hay plus barley or barley alone. They found no definite post-feeding increase with the all roughage diet when the blood was sampled at two hourly intervals. The plasma glucose levels in the present experiment do not reflect differences in ruminal TVFA concentration or ruminal propionic acid.

Urea: Plasma urea nitrogen concentrations were 125, 110 and 99 mg/l for the control silage, treated silage and dried grass respectively. Analysis of variance gave no significant evidence for differences between the diets. Vagher et al. (1973) quoted a range from 50 to 150 mg/l of blood urea nitrogen in calves. Lewis (1957) showed, for a variety of diets, that blood urea was relatively constant for a given diet, changes reflecting those in ruminal ammonia but being of a lower order of magnitude. Maximum blood urea concentration occurred some four hours after the maximum concentration of ruminal ammonia. Somers (1961) reported a range of blood urea concentrations from 196 to 276 mg/l for a diet of lucerne chaff, oat grain and cubes. Ruminal ammonia nitrogen concentration were 55 to 249 mg/l two and four hours after feeding, with blood values of 196 and 218 mg/l. Hawkins et al. (1970) for a silage diet, reported blood urea nitrogen values of 196 mg/l three hours after feeding and at the same time maximum ruminal ammonia concentration of 269 mg/l. Nishimuta et al. (1973) quoted plasma urea concentrations of 156 mg/l four hours after feeding and maximum ruminal ammonia concentration of 192 mg/l at two hours post-feeding.

These workers showed the effect of heat treatment of the diet in reducing plasma urea concentration as well as ruminal ammonia concentration. Plasma urea nitrogen levels, in the present investigation, are in keeping with the degree of heating involved in the production of the three dietary materials, but it is difficult to see how this effect could be mediated since ruminal ammonia concentrations did not reflect the heat treatments.

3B.

SILAGES MADE FROM GRASS OF LOW WATER SOLUBLE CARBOHYDRATE AND HIGH NITROGEN CONTENTS.

Nine sheep were used in the same experimental design as that shown in Experiments 2 and 3A (Tables 10 and 14). The sheep used are shown in Table 17.

Table 17

Designation of Sheep

Design No.	1	2	3	4	5	6	7	8	9
Sheep No.	443	435	409	434	447	449	437	448	414

Unfortunately, during period II the intakes of sheep 443 were very low and the sheep was removed from the trial. Sheep 68 replaced it in period III and sheep 680 was introduced in period III to replace sheep 443 in period II.

The treatments were:-

A. Control silage - 1213 g dry matter fed daily to each sheep.

B. Delayed sealed or treated silage - 1111 g dry matter fed daily to each sheep.

C. Dried grass - 828 g dry matter fed daily to each sheep.

The experimental procedures of the previous trial were used.

The outermost layers of the silage made after delayed sealing of the silo were non-uniform, varying in smell and texture. This material was discarded in favour of the inner core which was of a uniform smell and colour. The dry matter contents of the diets were 178, 187 and 920 g/kg and the pH values were 3.97, 5.30 and 6.12 for the control silage, treated silage and dried grass respectively. The composition of the silages and the dried grass are shown in Table 18.

Table 18

Composition of the Diets (g/kg dry matter)

	A (Control silage)	B (Treated silage)	C (Dried grass)
Crude protein	231	238	231
Crude fibre	267	319	250
MAD - fibre	326	378	291
Ash	113	135	121
Total nitrogen	37.0	38.0	37.0
Protein nitrogen	16.4	16.2	28.1
Water soluble nitrogen	20.6	21.8	8.9
Volatile nitrogen	4.0	8.0	-
Cellulose	300	319	277
Acetic acid	40	114	-
Propionic acid	2	14	-
Butyric acid	2	23	-
Lactic acid	105	tr	-
Succinic acid	tr	2	-
Water soluble carbohydrate	5	6	10.3
Ethanol	4.6	8.3	-

The treated silage contained, in addition, iso-butyric, iso-valeric and n-valeric acids in trace quantities.

RESULTS AND DISCUSSION

Nutritive Value

Mean daily dry matter intakes were 19.2, 28.1 and 32.9 g/kg $W^{0.75}$ for the control silage, treated silage and dried grass diets respectively. However, the amount of food given to the animals was restricted and the estimate of dried

grass intake does not reflect fully its intake potential. Harris and Raymond (1963) reported dry matter intakes by sheep of 79.5, 67.5 and 69.9 for S48 timothy, and 56.5, 56.3 and 54.2 g/kg $W^{0.73}$ for H1 ryegrass when conserved as barn dried hay, wilted silage and wilted silage made after forty-eight hours delay in sealing the silo. McDonald et al. (1960) reported sheep intakes of 739 g/day for fresh grass and 366 g/day for directly ensiled grass, compared with 715 g/day for the dried grass and 416 g/day for the directly ensiled material in the present investigation. Mean intakes for individual sheep are given in Appendix Table 71. Analysis of variance showed a significant difference in intake between the control silage and the other two diets.

Wilkins et al. (1971) claimed that the extensive degradation of silages of low fermentation quality was associated with low intake. However in the present trial, the treated silage with the lower fermentation quality was consumed in greater quantity. Brown and Radcliffe (1972) examined detailed chemical analyses, intake and in vivo digestibilities of twenty experimental silages with standardised wethers and suggested that silage intake was negatively correlated with the degree of fermentation. Moore et al. (1960) postulated that the low intake of silage could be related to some constituent formed during the ensiling process, probably in the nitrogen fraction. Baumgardt (1970) suggested that a contributing factor to the low intake of hay-crop silage could be the presence of soluble nitrogen compounds and Neumark et al. (1964) attributed it to the presence of amines, probably tryptamine. Large quantities of amines are generally associated with poorly preserved silages (Voss, 1966; Hughes, 1970). In the present investigation, the treated silage, badly preserved, with a volatile nitrogen content twice that of the control silage, was consumed in significantly greater amounts than the control.

Wilkins et al. (1971) reported a negative correlation between the acetic

acid content of seventy silages and voluntary consumption by sheep. Rumen infusions of acetic acid reduced dietary intake (Montgomery et al., 1963; Rook et al., 1963; Ulyatt, 1965; Weston, 1966 and Hutchinson and Wilkins, 1971). Montgomery et al. (1963) and Hutchinson and Wilkins (1971) found that neutralisation of the acid counteracted the depression but Bhattacharya and Warner (1968) did not confirm this effect. McLeod et al. (1970) reported increased intake when the free acid content of silage was reduced by bicarbonate addition. Hutchinson and Wilkins (1971) fed silages with concentrations of 20, 50 and 88 g/kg of acetic acid and found no significant difference in intake when the acids were neutralised to pH 4.1 with potassium hydroxide. Despite its higher acetic acid content the treated silage in the present investigation had a higher dry matter intake than the control. Ulyatt (1965) found propionic acid infusion into the rumen resulted in a small increase in intake, and Bhattacharya and Warner (1968) found that partial replacement of acetic acid by propionic acid in rumen infusion studies prevented depression in intake. The treated silage in this case had a higher propionic acid content than the control.

Gordon et al. (1961) found a poor correlation of lactic acid content with dry matter consumption of silage. Montgomery et al. (1963) infused lactic acid into the rumen and found only slightly decreased intakes. McLeod et al. (1970) found addition of lactic acid to silage decreased intake by as much as 13 g/kg $W^{0.75}$ when the original silage neutralised to pH 5.4 was reduced in pH to 4.8, 4.4 and 3.8 by addition of lactic acid. Senel and Owen (1966) found that a low addition of acetic and lactic acids did not alter the intake of silage but at a higher level of addition there was a beneficial effect on intake.

The higher intake of the treated silage was associated with a lower acidity than in the control and this accords with the published data on the effect of pH and neutralisation of acid on intake. The lactic acid content of

the control silage was very much higher than that of the treated and the effect on intake was as expected. However, it must be remembered that much of the proven associations of intake with chemical constituents have been established by regression analysis. Such analysis does not take into account that high pH may be associated with well preserved high dry matter silage, or silage which has undergone secondary fermentation. Equally, low lactic acid concentration may arise from restricted fermentation or as a result of destruction of lactic acid by clostridia. The two other characteristics which might be expected to have the greatest effect in reducing intake in the present investigation would be the extent of protein breakdown, and the acetic acid content. It is very surprising that the effect noted is the exact opposite. It would appear that either these two factors do not have such a drastic effect on intake as the published work would lead one to expect, or the characteristics of high pH and high propionic acid content have overridden their effect in the present instance.

Digestibility: Mean treatment values for the gross energy of digestible organic matter for the control silage, treated silage and dried grass were 24.4, 22.1 and 19.7 MJ/kg respectively. Analysis of variance showed a very highly significant difference between the three treatments. In the present trial the gross energies of dry matter for the control and treated silages, and the grass, were 21.5, 19.5 and 18.1 MJ/kg respectively, representing an increase of 0.18 during ensilage of the control, and 0.07 in the treated silage. McDonald et al. (1973) quoted a range of increases from 0.03 to 0.15 in silages with pH values ranging from 3.8 to 4.3. The relatively low gross energy of the treated silage in which considerable clostridial fermentation had occurred would suggest losses as gaseous hydrogen had taken place.

The mean digestibilities of dry matter, organic matter and nitrogen, and

metabolisable energy of the three diets are given in Table 19. The values for individual sheep are given in Appendix Table 71.

Table 19

Nutritive Values of the Three Diets

	A (Control silage)	B (Treated silage)	C (Dried grass)
Digestibility of dry matter	0.759	0.710	0.715
Digestibility of organic matter	0.788	0.755	0.753
Digestibility of nitrogen	0.835	0.794	0.806
Metabolisable energy (MJ/kg)	14.8	11.2	10.1

Statistical analyses using dry matter intake (g/day) as covariate showed a significant difference between the control silage and the other two treatments in digestibilities of dry matter, organic matter and nitrogen. Differences between the three treatments in metabolisable energy were also significant.

McDonald et al. (1960) in experiments to compare the effect of the consolidation of grass within the silo found decreases in dry matter, organic matter and nitrogen digestibilities of silages when the maximum temperature within the silo was 44° compared with the control maximum of 26°. In the present trial maximum temperatures for the treated and control silages were 40° and 25° respectively. Harris and Raymond (1963) found no difference in dry matter digestibilities of silages made with and without delayed sealing of wilted S48 timothy and H1 ryegrass. McDonald et al. (1966) confirmed their earlier work showing slight decreases in organic matter and nitrogen digestibilities with higher within silo temperatures when the maximum temperatures during ensilage of direct cut grass were 42° and 20°. However, in the same experiment when wilted

grass was used the nitrogen and organic matter digestibilities were slightly increased with the higher within silo temperature.

The higher digestibilities of the various fractions shown by the control silage is in keeping with published work. The lower values for the treated silage agree with the work of McDonald et al. (1960, 1966) but not with that of Harris and Raymond (1963). It would appear that the heating which occurred in the production of the treated silage may have been responsible for the lowering of the digestibilities, but it is difficult to reconcile this with the relatively high figures for the dried grass which received the most intensive heat treatment. The metabolisable energy values are in the order to be expected of the gross energy and digestibility figures.

Rumen Characteristics

The values for rumen fermentation products have been adjusted for level of intake effect by analysis of covariance with dry matter intake (g/day) as the covariate. The adjustments of the values were small. For the control silage, with the lowest intake, the adjustment involved a very slight lowering of rumen pH and increase of TVFA concentration and negligible changes in the molar proportions of acetic and propionic acids. Statistical comparison of the fermentation curves has been made on the basis of the first slope, considered to be from the pre-feeding to the one hour post-feeding sample, the maximum of parabola for time of attainment and concentration, and the second slope, from the third hour post-feeding to the next pre-feeding sample.

The curves illustrating changes in ruminal fermentation characteristics with time of sampling after feeding, for each treatment, are based on adjusted mean values for nine sheep. Compositional data for the rumen contents of

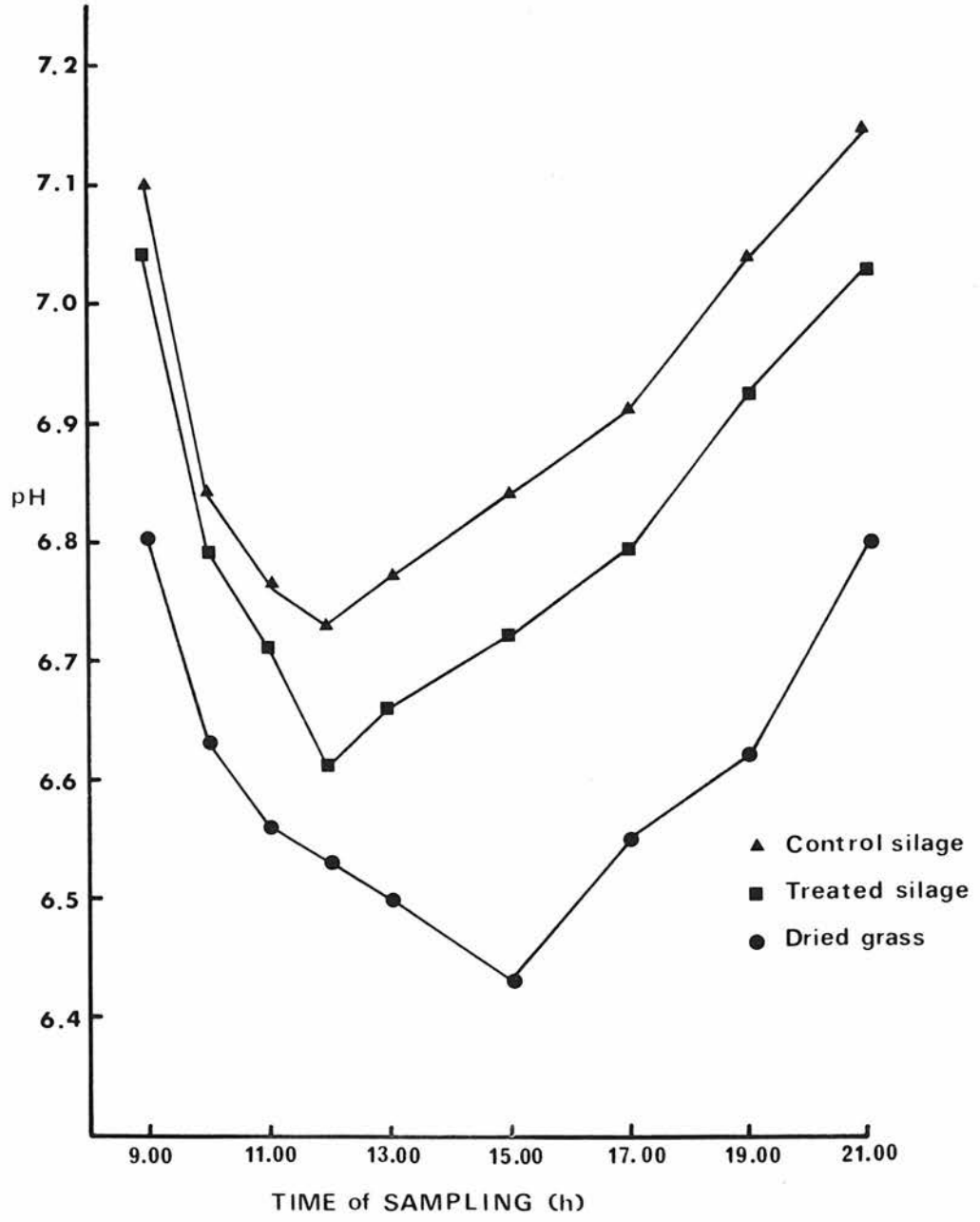

Fig. 34 RUMINAL pH VALUES

individual sheep fed each diet are given in Appendix Tables 72 to 98.

pH and TVFA Concentrations: Fig. 34 shows the curves of rumen pH values for the three treatments. The pH values are lower at all stages for the dried grass treatment than for either of the silages. Analysis of covariance gave no significant evidence for differences between the silages at any sampling stage. Differences between the dried grass and the other two treatments at 09.00, 10.00, 15.00, 17.00, 19.00 and 21.00 h. were significant. Differences between the dried grass and the control silage at 13.00 h. were significant. Analysis of variance gave no significant evidence for differences between the treatments in the first or second slopes, the pH value or the time of minimum of parabola. However, the latter value was much higher for the dried grass at five hours than for the silages at three hours post-feeding.

The lower ruminal pH values for the dried grass compared with the silage diets in the present work reflect the higher sugar content of the grass and is in agreement with the results reported by Svensson and Palsson (1966) who found lower maximum and minimum values for hay compared with silage and with Anderson and Jackson (1971) for first cut hay and silage. However, a comparison of third cut hay and silage by the latter authors showed no difference in ruminal pH values and, with a second cut, the ruminal pH values were lower with silage, confirming the values of 6.21 for hay and 6.01 for silage (late cut), quoted by Bath and Rook (1965). The rumen pH patterns with the silage diets in the present work are similar to that reported by Devuyst et al. (1968) for a pelleted hay diet supplemented with starch when maximum pH values of 7.0 were at pre-feeding and minimum values of 6.69 at two and a half hours post-feeding. Schmekel (1967) reported a range from 7.04 to 6.83 for a silage diet with a

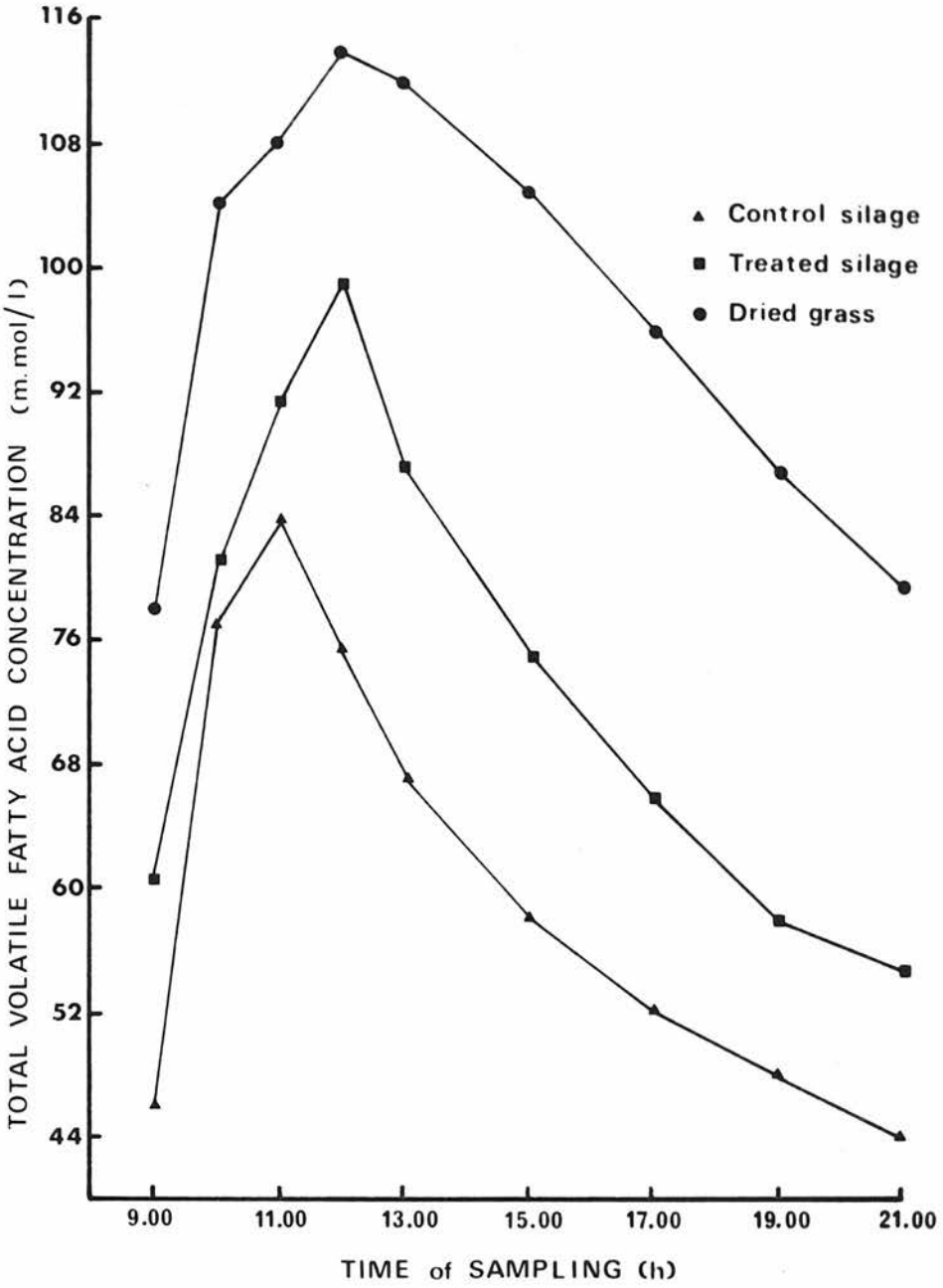

Fig. 35 RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATIONS

minimum value at three hours post-feeding. Steger et al. (1970) reported a similar pattern, and a range from 7.1 to 6.5 for a maize silage diet. This is similar to that in the present trial for both silage diets.

The ruminal TVFA concentration curves for the three treatments at the nine sampling stages are given in Fig. 35. The concentration is greater at all post-feeding sampling stages for the treated compared with the control silage, although only at 13.00 h. is the difference significant. The concentration of TVFA is greater for the dried grass than either of the silages at all the sampling stages. Analysis of variance showed the differences between the dried grass and the silages to be significant at all stages except at 12.00 h. Analysis of variance gave no significant evidence for differences between the treatments in the first or second slopes or the time of maximum of parabola, but showed a significant difference between the dried grass and the control silage in the maximum of parabola for TVFA concentration in the rumen. The slightly higher ruminal TVFA concentration of 98 m mol/l for the treated compared with 83 m mol/l for the control silage and its later post feeding time of maximum concentration may be due to its higher fibre content of 378 g/kg compared with 326 g/kg for the control, since Raun et al. (1962) and Templeton and Dyer (1967) reported increased TVFA concentration when the hay inclusion in a mixed concentrate roughage diet was increased from 0.2 to 0.5 parts. The latter workers quote an increase in TVFA concentration from 107 to 137 m mol/l with an increase in crude fibre from 105 to 191 g/kg.

In comparing hay with silage Bath and Rook (1965) found a ruminal TVFA concentration of 124 m mol/l for late cut silage compared with 95 for hay, while Anderson and Jackson (1971) found lower ruminal TVFA values for silages than hay made from first, second or third cut grass, which agrees with the lower TVFA concentration for the silage diets compared with the dried grass in the present

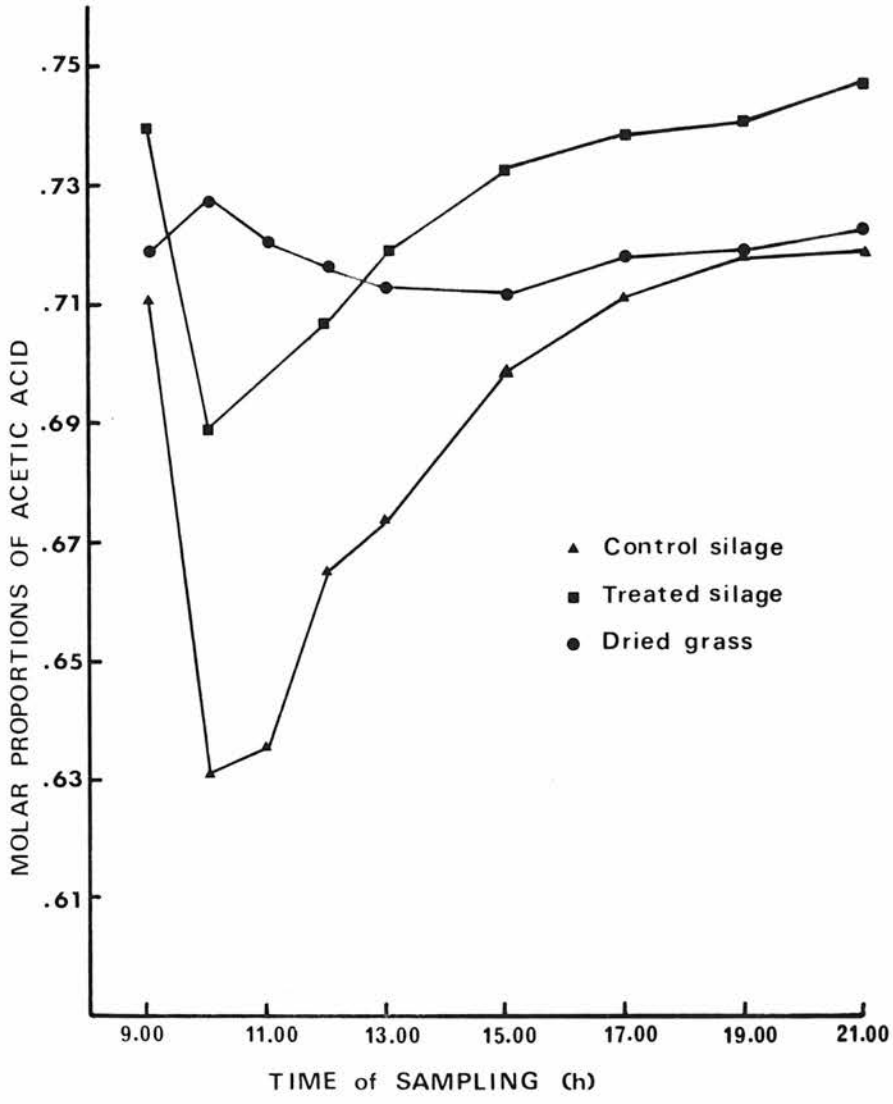

Fig. 36 RUMINAL MOLAR PROPORTIONS OF ACETIC ACID

investigation. El Shazly (1952) sampled rumen contents before and after feeding silage and reported TVFA concentrations of 55 to 110 m mol/l similar to those of 55 to 99 m mol/l for the treated silage but his values for a dried grass of 94 to 135 m mol/l were higher than those in the present trial.

Similar ruminal TVFA concentration patterns were reported by Devuyst et al. (1968) for a diet of lucerne pellets. They reported minimum values of 60 m mol/l at pre-feeding and maximum of 110 m mol/l at two and a half hours post-feeding, and a range of 72 to 116 m mol/l when the pellets were supplemented with starch. The patterns and values were also similar to those found by Steger et al. (1970) who reported minimum pre-feeding concentrations of 63 and 90 m mol/l and maximum concentrations of 93 and 115 m mol/l between one and three hours and at three and a half hours post-feeding respectively for diets of maize silage and hay.

Major Acids: Fig. 36 shows the molar proportions of acetic acid in the rumen contents at the nine sampling stages for the three diets. The molar proportions of acetic acid were lower for the control silage than for the other two diets at all sampling stages. Analysis of variance showed a significant difference between the treated silage and the dried grass at 10.00 h. Differences between the control silage and the other two treatments at 10.00, 11.00, 12.00 and 13.00 h., and between the silages at 09.00 and 15.00 h. were significant. The curve pattern of the molar proportions of acetic acid in the rumen when the dried grass was fed was different from those with the silage diets. There was no rapid decline to minimum values but a slight increase to a maximum value, and analysis of variance showed a significant difference between the dried grass and the silages when the first slopes were compared. At the minimum of parabola (concentration) there were significant differences between the dried grass and

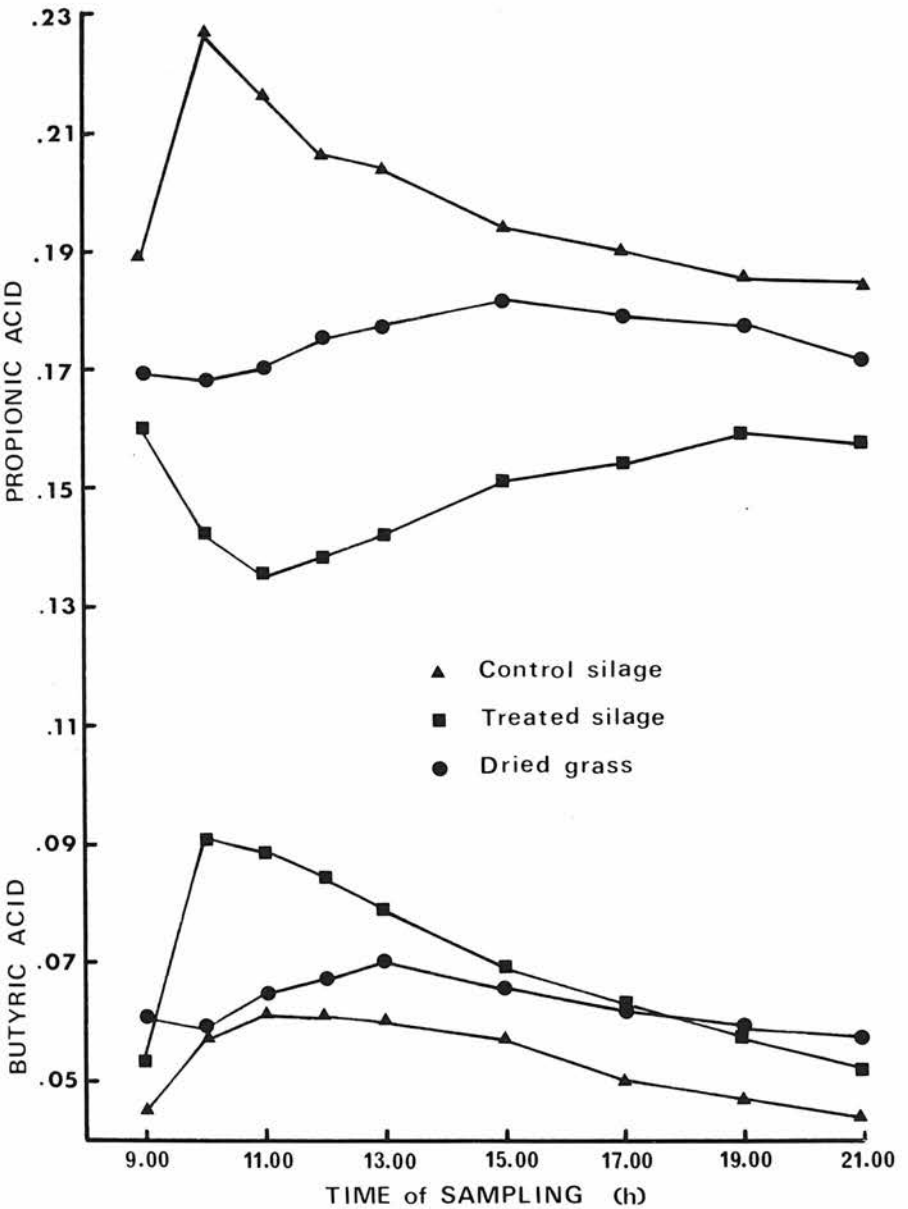

Fig. 37 RUMINAL MOLAR PROPORTIONS OF PROPIONIC AND BUTYRIC ACIDS

the control silage and between the silages. Differences between the control silage and the dried grass in the second slope were significant.

Fig. 37 shows the ruminal molar proportions of propionic acid over the nine sampling stages. There is a different pattern curve with each diet, the biggest difference being between the silages. Analysis of variance showed significant differences between the silages at all the sampling stages. Differences between the dried grass and the silages at 10.00, 11.00, 12.00 and 13.00 h., between the dried grass and the control silage at 09.00 h. and between the dried grass and the treated silage at 15.00, 17.00 and 19.00 h. were significant. Differences between the control silage and the other two treatments in the first slope were significant as were those between the maxima of parabola (concentration) for the three treatments. Differences between the treated silage and the other two diets in the second slope were significant. Fig. 37 also shows the molar proportions of butyric acid in the rumen contents. The proportions were higher at all except pre-feeding sampling times, when the treated silage diet was fed. Analysis of variance showed that differences between the silages at 10.00, 11.00, 12.00 and 13.00 h. were significant. The differences between the dried grass and the treated silage at 10.00, 11.00 and 12.00 h. were significant as was that between the dried grass and the control silage at 09.00 h. Analysis of variance showed differences between the first slopes and between the maxima of parabola (concentration) for the treated silage and the other two diets were significant as was that between the treated silage and the dried grass in the second slope.

El Shazly (1952) reported a range of ruminal acetic acid values from 0.73 to 0.67 for a silage diet, compared with 0.72 to 0.64 for the control silage in the present investigation. The range for the treated silage of 0.75 to 0.69 was similar to that of 0.74 to 0.70 given by Balch and Rowland (1957) for a hay

plus concentrate diet.

The curve pattern of acetic acid and the values in the rumen contents for the control silage in the present work are similar to those of Puech et al. (1968) with maximum pre-feeding values of 0.72 and minimum of 0.63 at two hours post-feeding for a directly ensiled silage diet and Steger et al. (1970) found a similar pattern but not of values with a diet of clover-grass silage plus dried green fodder. The same authors (1970) reported a maximum pre-feeding value of 0.75 and a minimum three hour post-feeding value of 0.70 for a maize silage diet, which was similar to the treated silage in the present experiment.

The similar patterns, though at different levels of the molar proportions of acetic acid in rumen contents when both silage diets were fed, confirm the views of Bath and Rook (1965), Kaufmann and Rohr (1967) and Anderson and Jackson (1971), who all showed a high correlation between the fibre content of the diet and acetic acid proportions in the rumen juice.

The very different pattern of the proportions of acetic acid in the rumen contents after feeding the dried grass diet compared with the silages was similar to that found by Puech et al. (1968) for a hay diet with pre-feeding values of about 0.70 and maximum values of about 0.71 at two hours post-feeding and to those of Devuyst et al. (1968) for a pelleted lucerne hay when the molar proportions of acetic acid in the rumen was maximal at two hours post-feeding and the range of values was very narrow. This narrow range of values for the dried grass diet in the present work recalls those obtained with animals grazing pasture herbage (Balch and Rowland, 1957).

In the present work the small differences in the molar proportions of propionic acid in the rumen contents during the post-feeding period when the diet was dried grass reflect the absence of differences in acetic acid proportions. This was also shown by Puech et al. (1968) for a hay diet. Balch and

Rowland (1957) for an arable silage diet, Puech et al. (1968), for a hay diet, Devuyst et al. (1968), for a pelleted lucerne diet and Steger et al. (1970), for a maize silage diet, reported a similar narrow range of molar proportions of propionic acid to those for the dried grass diet in the present investigation.

The difference in pattern and in the values of propionic acid, when the silage diets are compared, is remarkable. The range of values for the control silage was similar to those found by Balch and Rowland (1957) for a hay plus concentrate diet, and the pattern was similar to that of Puech et al. (1968), with a minimum pre-feeding value of 0.17 and a maximum of 0.22 at two hours post-feeding, when silage was fed. The decrease in the proportions of propionic acid after feeding the treated silage was unusual. Briggs et al. (1957) reported a decrease in ruminal propionic acid proportions in some sheep after feeding diets high in starch and casein, and Steger et al. (1970) showed with a maize silage diet a pre-feeding proportion of propionic acid of 0.175 which declined to 0.160 thirty minutes after feeding and then rose to a maximum of 0.185 at three hours post-feeding. Neither Balch and Rowland (1957) for a wide variety of diets, nor El Shazly (1952) for diets of silage, fresh or dried grass, reported values as low as 0.135 when quoting a range of values of ruminal propionic acid proportions. However, Anderson and Jackson (1971) quoted 0.127 for an average post-feeding value of rumen contents from one sheep on a low dry matter silage. The silage was similar to that in the present work in that it was made from an autumn cut, but the herbage was consolidated in the silo. Unfortunately, no in-silo temperatures were recorded (Anderson and Jackson, 1970).

There was very little variation in the molar proportions of butyric acid in the present trial when the dried grass was fed, as might be expected from a consideration of the acetic and propionic acid curves. The values are similar to the 0.066 for a hay diet quoted by Bath and Rook (1965). El Shazly (1952)

quoted a pre- and post-feeding difference of 0.003 for a dried grass diet but his values were higher than those in the present trial.

The low values of butyric acid found in the rumen contents with the silage diets in the present work, particularly the 0.044 to 0.061 for the control silage, were considerably less than those quoted for silage diets by Bath and Rock (1965), by Schmekel (1967) and by Anderson and Jackson (1971). From twenty results the latter authors quoted one value of ruminal butyric acid below the maximum value for the control silage diet in the present investigation. Similar very low butyric acid proportions were found in rumen contents by Reid et al. (1957) for a diet of crushed oats, wheaten chaff, lucerne chaff and cracked maize. Balch and Rowland (1957) quoted values of 0.050 to 0.074 for an arable silage diet.

Patterns of ruminal butyric acid proportions similar to those obtained for the treated silage in this experiment were obtained by Puech et al. (1968), who quoted a pre-feeding minimum of 0.087 and a maximum of 0.125 at two hours post-feeding, for a silage diet. Steger et al. (1970) found a slight post-feeding increase from 0.075 to 0.090 at one hour post-feeding for a maize silage diet, which is similar to the changes noted for the control silage diet in this experiment.

Although not unique, the changes in the relative proportions of the major volatile fatty acids following feeding in the present work are unusual in that there was an unexpected slight rise in acetic acid on the dried grass diet and correspondingly little change in propionic and butyric acid levels. Although both the control and treated silages showed the expected post-feeding fall in acetic acid, that of the treated silage was balanced by a rise in butyric acid instead of propionic acid which fell slightly.

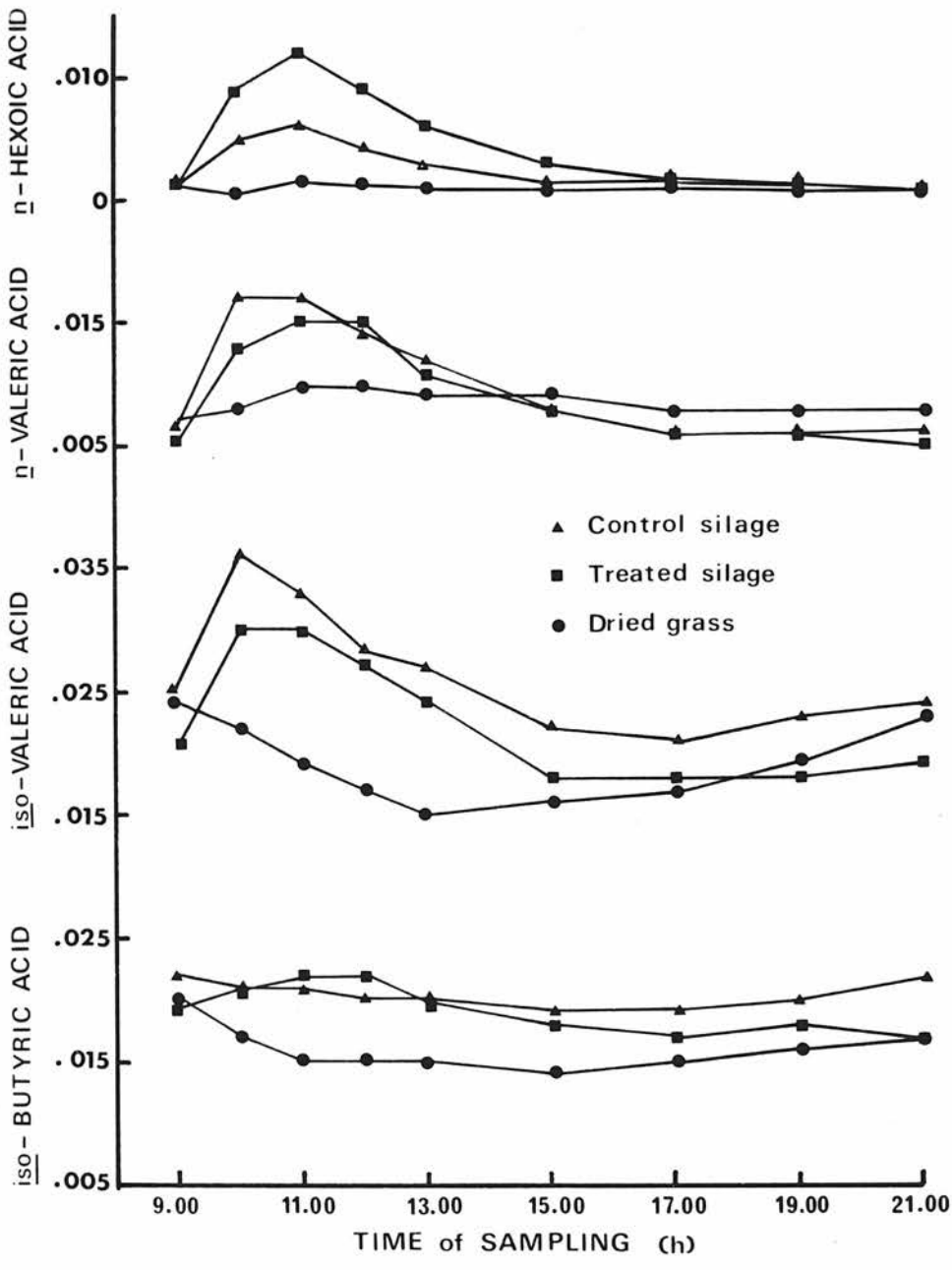

Fig. 38 RUMINAL MOLAR PROPORTIONS OF iso-BUTYRIC, iso-VALERIC, n-VALERIC AND n-HEXOIC ACIDS

Minor Acids: Fig. 38 shows the molar proportions of iso-butyric, n-valeric, iso-valeric and n-hexoic acids in the rumen contents sampled at nine stages after giving the three diets. The values for the dried grass diet are different from the silage diets in pattern and level.

Analysis of variance of the treatment values for iso-butyric gave no significant evidence for differences between the silages at any sampling stage. Differences between the dried grass and the silages at 10.00, 11.00, 12.00, 13.00 and 15.00 h. were significant. Comparison of the first slopes showed significant differences between the treated silage and the other two diets. Differences between the dried grass and the other two treatments in the maximum of parabola were significant, and in the second slope differences between the silages, and between the dried grass and the control silage were significant. Analysis of variance showed the differences between the ruminal proportions of iso-valeric acid for the dried grass and the silages at 10.00, 11.00, 12.00 and 13.00 h., and between the silages at 10.00 h. were significant. Differences between the dried grass and the silages in the first and second slopes and the maximum of parabola were significant. Differences in the ruminal proportions of n-valeric acid between the dried grass and the silages at 10.00, 11.00 and 12.00 h. were significant as were those between the dried grass and the treated silage at 09.00, 19.00 and 21.00 h. Differences between the silages at 09.00, 10.00 and 11.00 h. were also significant. Differences between the dried grass and the silages in the first and second slopes, and in the maximum of parabola were significant. Analysis of variance of the treatment values at the nine sampling stages for n-hexoic acid showed differences between the three treatments at 10.00, 11.00 and 12.00 h., and between the treated silage and the other two diets at 13.00 and 15.00 h. were significant. Differences between the three treatments in the first and second slopes and the maximum of parabola were

significant.

The higher values of ruminal iso-butyric acid found in the present work for the silages compared with the dried grass diets confirm the findings of Bath and Rook (1965) and El Shazly (1952). The change following feeding of the control silage is similar to that found by the latter worker. The changes in the ruminal proportions of iso-valeric acid, following feeding of the dried grass in the present trial, are similar to those reported by El Shazly (1952) for dried grass diets, but for the silage diets are opposite to those of Schmekel (1967) for silage. The higher values for the silages compared with the dried grass in the present work agree with the findings of El Shazly (1952) for dried grass and Fenner et al. (1970) for hay compared with silages.

The higher values of n-valeric acid in the rumen contents for the silage diets compared with the dried grass agree with the reports of Bath and Rook (1965) and Anderson and Jackson (1971) who compared silages with hays. Fenner et al. (1970) found higher ruminal n-valeric acid proportions for silage compared with hay and they and Schmekel (1967), with a silage diet, reported an increase in ruminal n-valeric acid after feeding, which was maximal at three hours post-feeding, a pattern which was similar to that found for silages in the present work. Fenner et al. (1970) reported a sevenfold increase in ruminal n-caproic acid when maize silage replaced hay. The pattern was similar to those found in the present trial with a fairly sharp increase to maxima two to three hours after feeding.

The higher values for the iso-butyric and iso-valeric acids in the rumen contents when the silage diets were compared with the dried grass in the present investigation reflect the in-silo proteolytic breakdown and hence the increased formation in the rumen of the branched chain acids from amino acids. The dichotomy between the silages and the dried grass in the ruminal proportions of

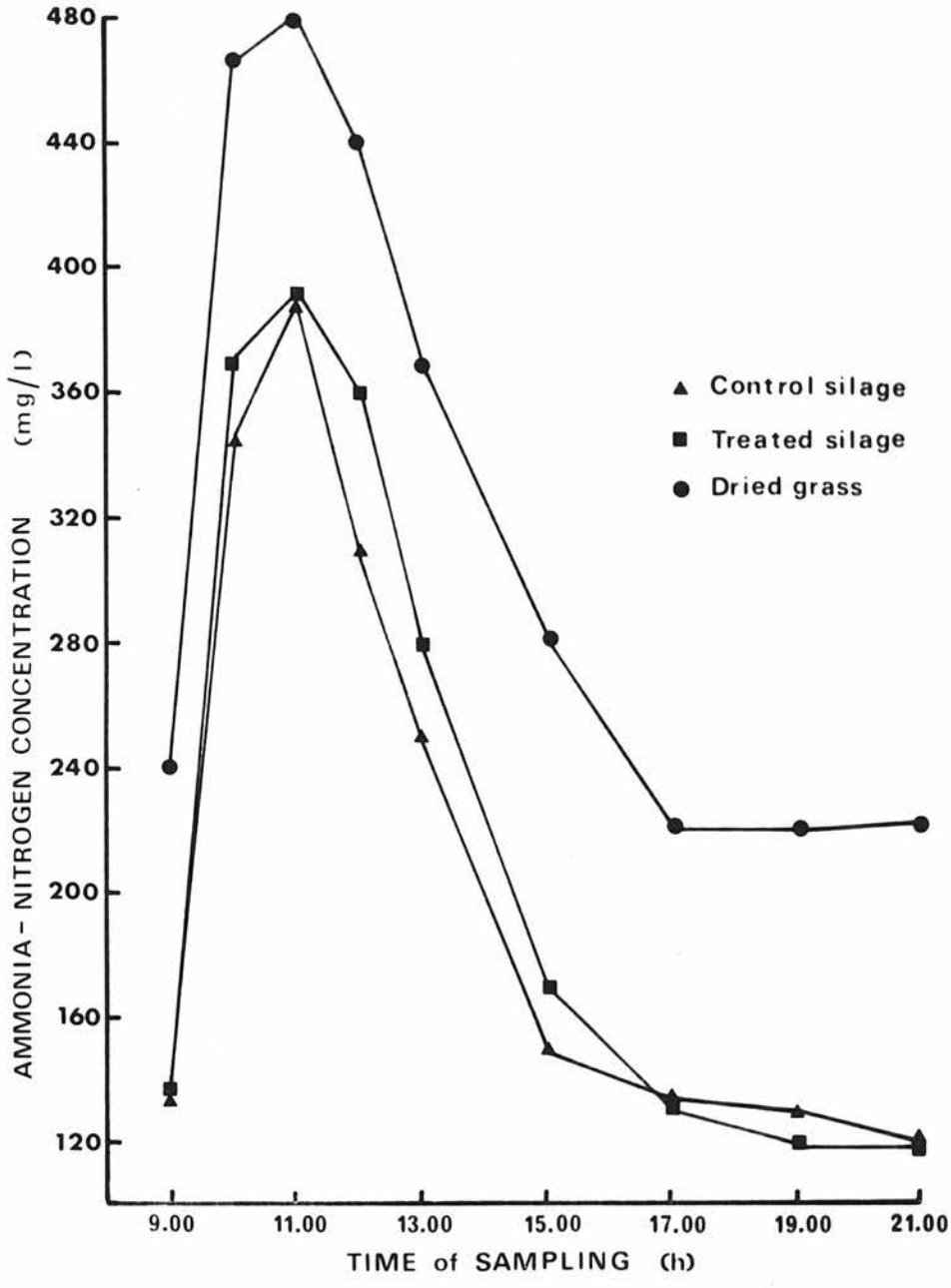

Fig. 39 RUMINAL AMMONIA CONCENTRATIONS

n-hexoic acid does not reflect this proteolysis since this acid is produced from lactic acid and not deamination of amino acids.

Fig. 39 shows the ammonia concentration in the rumen contents of the sheep fed the three diets. Analysis of variance gave no significant evidence for differences between the silage diets at any of the sampling stages. Differences between the silages and the dried grass were significant at all stages except 12.00 h. when the difference between the dried grass and the control silage only was significant. The first or second slopes of the curves were not significantly different for any of the diets but the differences between the maxima of parabola for the silages and the dried grass were significant. Chalmers (1963) reported higher maximum rumen ammonia concentrations with silage compared with a dried grass diet. In the present work, the maximum concentration of ammonia in the rumen contents was higher with the dried grass diet, minimum pre-feeding values were also higher and the post-feeding increases in ammonia nitrogen concentration were 271, 273 and 258 mg/l for the control silage, treated silage and dried grass diets respectively. The difference in composition, particularly in the nitrogen fraction of the diets, may be responsible for the higher minimum pre-feeding values for the dried grass compared with the silage diets since the treatment could have resulted in a nitrogen fraction more resistant to breakdown. Davis and Stallcup (1967) reported higher maximum and minimum values of ruminal ammonia with soyabean meal compared with raw soyabean. Sutton and Vetter (1971) reported higher pre-feeding minimum and post-feeding maximum concentrations for a hay diet compared with a silage diet. The values and pattern of ruminal ammonia concentration in the present investigation with the silage diets are similar to those reported by Durand et al. (1968) with pre-feeding minimum of 130 mg/l and maximum value at two hours post-feeding of 420 mg/l for a diet of lucerne silage.

Blood Characteristics

Blood pH, plasma glucose and urea values for individual sheep are given in Appendix Table 99.

pH: Blood pH values were 7.32, 7.35 and 7.36 for the control silage, treated silage and dried grass diets respectively. Analysis of variance gave no significant evidence for differences between the treatments. Vagher et al. (1973) quoted an average pH value of 7.418 with a range from 7.375 to 7.461 for thirty-one calves. Their minimum value was higher than those for the sheep in the present work. The pH values reported here are not in accord with the rumen pH values and confirm the lack of influence of rumen pH on blood pH. This might be expected from the massive buffering capacity of blood.

Glucose: Plasma glucose concentrations were 552, 571 and 594 mg/l for the control silage, treated silage and dried grass diets respectively. Analysis of variance showed the differences between the control silage and the dried grass were significant.

Annison et al. (1957) and Armstrong and Blaxter (1957a) for sheep, Schultz and Smith (1951) for goats, and Waldo and Schultz (1960) and Storry and Rook (1965b) for cows, reported increased plasma glucose levels with ruminal infusions of propionic acid. Annison et al. (1963) suggested that absorbed propionic acid was the source of 0.30 of the glucose synthesised while Leng et al. (1967) and Judson et al. (1968) gave estimates of 0.54 and 0.46 respectively. Bensadoun et al. (1962) suggested that gluconeogenesis from precursors other than propionic acid was important. Krebs (1964) calculated that 100 g of protein could yield 50 to 60 g of glucose. Lindsay (1970) from available data suggested that the

contribution of protein to glucose synthesis was low and reported little stimulation in glucose production from perfused sheep liver by addition of casein hydrolysate. Ford (1965) suggested that the greater rate at which glucose entered the circulation of ruminants when the diet was spring grass compared with a hay plus oats diet was due to the higher intake of protein.

In the present investigation levels of blood glucose were not linked with the concentration or the proportion of propionic acid in rumen contents. The level of protein nitrogen in the dried grass was higher than either of the silages and this could account for the higher level of plasma glucose with this diet. This does not, however, account for the difference between the two silages.

Urea: Plasma urea nitrogen concentrations were 217, 241 and 266 mg/l for the control silage, treated silage and dried grass respectively. Analysis of variance gave no significant evidence for differences between the treatments. Abou Akkada and Osman (1967) reported a significant correlation between blood urea and ruminal ammonia when the correlation was based on roughages. The results of the present investigation with the higher ruminal ammonia and plasma urea concentrations for the dried grass diet compared with the silages agree with their report. Sutton and Vetter (1971) also found a similar relationship, and their values for hay were higher than those for silage. Their values for blood urea concentration at three hours post-feeding were 265 and 205 mg/l for hay and silage respectively, which are similar to the values for the dried grass and control silage in the present investigation. Their corresponding ruminal ammonia concentrations at one hour post-feeding were 380 and 320 mg/l for the hay and silage diets respectively. Durand et al. (1968) quoted a ruminal ammonia concentration of 420 mg/l two hours after feeding lucerne silage and a blood urea nitrogen concentration of 250 mg/l three hours after feeding, which is similar to the values recorded for the treated

silage. The different blood urea nitrogen concentrations recorded for the two silages which had very similar ruminal ammonia nitrogen levels shows the association between these two parameters to be incomplete.

GENERAL DISCUSSION OF EXPERIMENTS 3A and 3B.

Eight of the nine sheep were common to both experiments but only a superficial comparison seemed justified owing to the time interval between them, and the fact that the values for digestibility and rumen fermentation products had been adjusted within each experiment to allow for the effect of intake.

Comparison of the Silages

Dry matter intake varied between the silages, those of the treated silages being higher than the corresponding control silage. In agreement with the published work the intake of dry matter appeared to be a reflection of the end products of the fermentation in-silo and not upon any one particular component of silage. Low water soluble carbohydrate content, low pH, high nitrogen both volatile and water soluble, and high acid concentrations were associated with the lowest intake. Conversely high water soluble carbohydrate content, high pH, low organic acid and low nitrogen contents were associated with the highest intake. The delayed sealing of the silo had about the same effect in both silages.

Delayed sealing of the silo decreased digestibility of dry matter, organic matter and nitrogen and as would be expected gave silages of lower gross energy than the control, the lowering being greater in the case of the third cut material. In spite of this reduction, intakes of metabolisable energy were higher for the delayed sealed materials owing to their higher dry matter intakes. Thus for a 50 kg sheep intakes of metabolisable energy can be calculated as 8.4 and 6.1 MJ for the delayed sealed and control silage made from first cut material and 5.9 and 5.0 MJ for those made from the third cut grass. Thus in both cases delayed sealing conferred definite advantages in terms of production potential and, as expected, the first cut material was best.

Delayed sealing *per se* had little or no effect on rumen pH, TVFA concentration or the molar proportions of butyric acid, although the third cut delayed sealed product tended to show higher values for the latter. The ranges of variation in these parameters was greater with the first cut material.

The effect of delayed sealing on the ruminal molar proportions of acetic and propionic acids was different for the first and third cuts. The treated first cut had a lower ratio of acetic to propionic acid than the control silage, while with third cut material the treated had a higher ratio. It could thus be expected that the efficiency of utilization of metabolisable energy would be higher for first than for third cut material. It is interesting that with the third cut control silage and the first cut treated silage the ratio of acetic to propionic acids was less than 3.0, the critical level below which Armstrong and Prescott (1971) suggested that milk fat content would be lowered.

Ruminal *iso*-butyric and *iso*-valeric acids were higher for the third cut silages with the higher nitrogen content compared with the first cut. Kaufmann and Rohr (1967) reported higher values of iso-acids with higher dietary protein content. Ruminal *n*-valeric acid was slightly higher for the control silage and *n*-hexoic acid slightly higher for the treated silage with third compared with first cut grass. Delayed sealing did not appear to have any real effect on the proportions of the minor volatile fatty acids of the rumen.

Although with the first cut material averages of 21 and 14 g of lactic acid were ingested at each meal from the control and delayed sealed silages respectively and with third cut material trace quantities for the treated and 21 g for the control silage, no lactic acid was detected in rumen contents one hour after feeding.

Ruminal ammonia concentrations were lower for the early cut silages with the lower nitrogen contents. Delayed sealing of the silo had no apparent effect on ammonia concentration in either experiment.

Plasma glucose levels were slightly lower for the first cut compared with the third cut silage. Delayed sealing of the silo increased the value slightly in both experiments.

Plasma urea levels were higher for the third cut silages with the higher nitrogen contents and the higher ruminal ammonia concentrations. The higher values confirm the results of Voigt et al. (1969) who reported increased plasma urea concentrations in cows at grass given a high nitrogen fertiliser application.

Overall it would appear that delayed sealing confers advantages from the point of view of production potential owing mainly to the improved intake which accrues. It is noteworthy that although the treated third cut silage would have been classed as the poorest silage in most laboratory evaluation estimates, it is, according to these results, considerably better than the control third cut and as good as the first cut control.

Comparison of the Dried Grasses

Intake of the first cut material was higher, which is in keeping with its higher water soluble carbohydrate content, but may also reflect the differences in the quality and composition of the nitrogen fraction.

The digestibility of nitrogen was markedly higher for the third compared with first cut grass. On the other hand digestibilities of organic matter and dry matter were higher for the first cut material, but not so the metabolisable energy owing to the lower gross energy of digestible organic matter of the first cut material. The higher protein content would account for this.

Ruminal pH and TVFA concentrations indicate a more active rumen fermentation following feeding in the case of the first cut grass and this fermentation was more propionate orientated and gave higher molar proportions of butyric acid.

As would be expected from its higher nitrogen content the third cut material gave higher proportions of ruminal iso-butyric and iso-valeric acids and higher concentrations of ruminal ammonia.

The composition of the third cut grass is typical of much autumn grass which is generally held to give disappointing results in practice compared with spring grass. Despite its higher fibre content the third cut grass in the present experiment had virtually the same metabolisable energy as the first cut. However, it is likely that this energy would be less efficiently utilised owing to its high nitrogen content and the lower proportion of propionic acid in the end products of rumen fermentation. Probably, though, the major factor affecting the production potential of the grasses is the different intakes which can be achieved when they are fed.

3C.

A COMPARISON OF RUMEN FERMENTATION PATTERNS WITH FRESH AND DRIED GRASS DIETS.

Grass of High Water Soluble Carbohydrate and Low Nitrogen Contents (Experiment 3A).

Grass of Low Water Soluble Carbohydrate and High Nitrogen Contents (Experiment 3B).

INTRODUCTION

In the previous experiments (Experiments 3A and 3B) it was not possible to feed cut grass and silages made from the same source. Grass, artificially dried at low temperature was used as the control diet. Jentsch et al. (1972) concluded that gentle drying of green fodder did not change its chemical composition, its gross energy, nor the digestibility of the energy, or the proximate constituents. This experiment was an attempt to compare the rumen fermentation patterns when both fresh and dried grasses were fed. The dried grasses used were those of Experiments 3A and 3B and the rumen fermentation patterns of these have been presented earlier in this section.

EXPERIMENTAL

Fresh grass was cut daily from the same source as that to be dried or ensiled, and was fed to five sheep in two discrete meals per day, over a fourteen day period prior to cutting for conservation. The feeding regime was the same as for silage diets. Rumen contents were sampled one day before and one day after ensiling. The composition of the fresh grasses fed during the last seven days are shown in Appendix Tables 100 and 101.

In the first trial 4.5 kg of the fresh grass of Experiment 3A and in the second between 4.5 and 5.5 kg, depending on the assessed dry matter content, of the fresh grass of Experiment 3B was offered daily to each sheep.

RESULTS AND DISCUSSION

Nutritive Value

Intake: Mean daily intakes of dry matter for the four diets are shown in

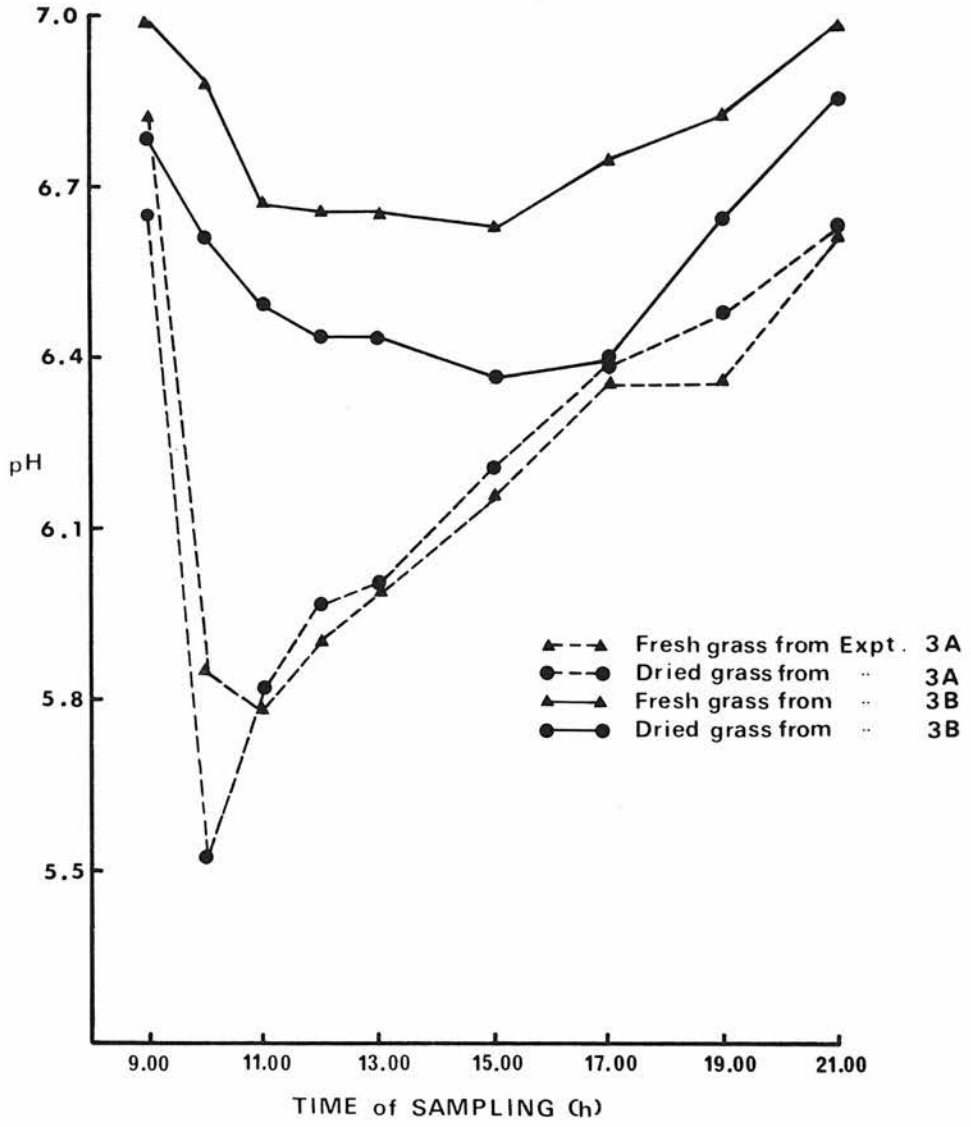

Fig.40 RUMINAL pH VALUES

Table 20.

Table 20

Daily Dry Matter Intake of Grasses ($g/kg\ W^{0.75}$).

	Fresh	Dried
Grass of Experiment 3A	41.7	40.6
Grass of Experiment 3B	26.4	36.2

These figures are not a true reflection of the absolute intake potential of these diets owing to restrictions on the amount provided and the limited access time allowed. The low dry matter content of the fresh grass of Experiment 3B combined with the restricted time of access for eating may have contributed to the low dry matter intake of this diet and, additionally, the individual intakes of one sheep were exceptionally low. Heaney et al. (1966) have also reported reduced intakes of fresh, autumn grass compared with dried material.

Rumen Characteristics

Statistical comparison of the values of rumen fermentation products at pre-feeding and at one, two, four and six hours post-feeding has been made with four sheep which were fed both the fresh and dried grass of Experiment 3A, and with three sheep which were fed the fresh and dried grass of Experiment 3B. The composition of the rumen contents sampled at the nine intervals, for individual sheep on both fresh grass diets, are given in Appendix Tables 102 to 108.

pH and TVFA Concentration: pH values for the rumen contents at the nine sampling stages for the fresh and dried grasses are shown in Fig. 40. The pH was lower for

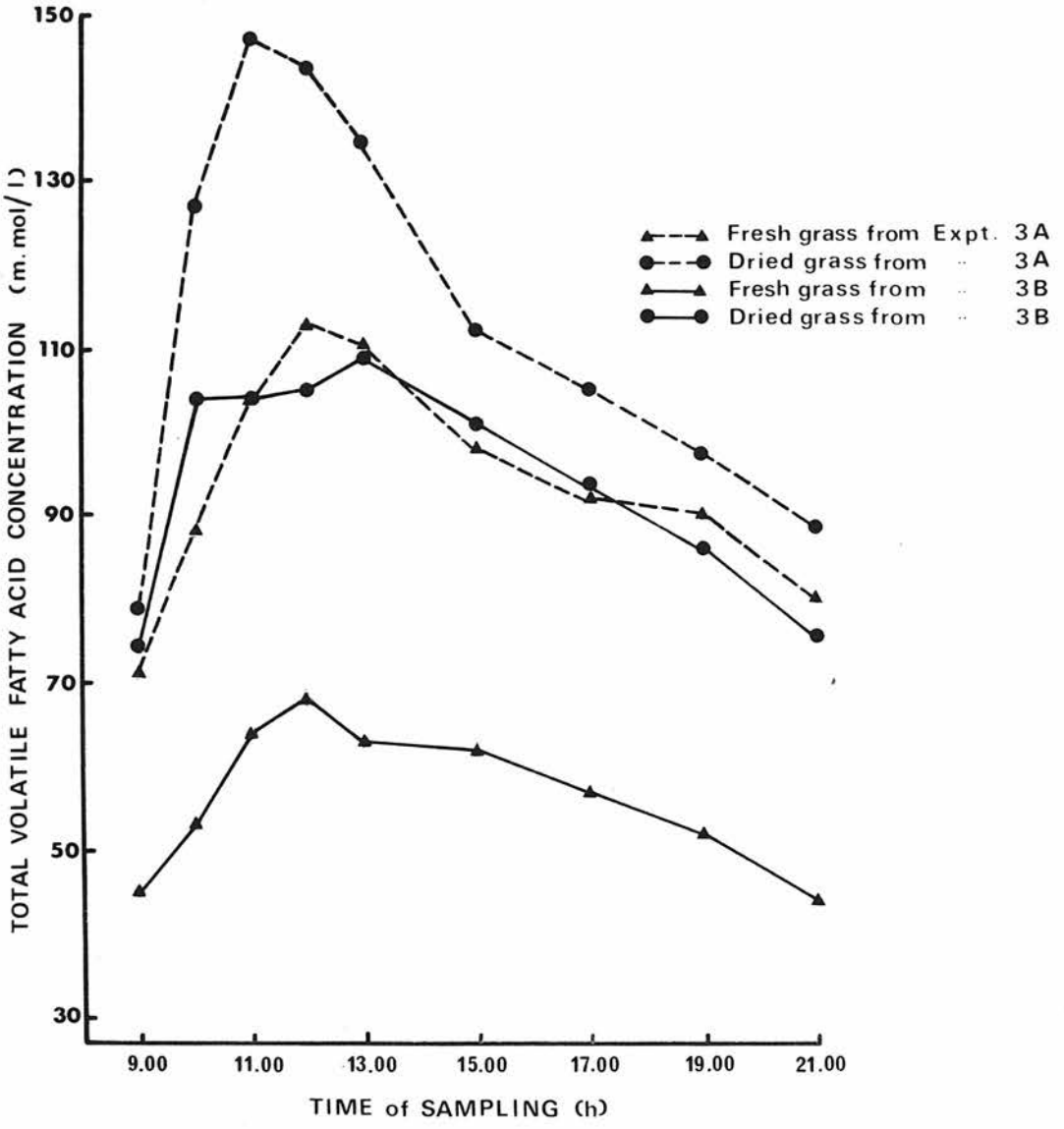

Fig.41 RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATION

the dried compared with the fresh grass at all sampling stages in Experiment 3B, but only at 09.00 and 10.00 h. in Experiment 3A. Analysis of variance gave no significant evidence for differences between the fresh and dried grass at any of the five sampling stages. Christian and Williams (1957) also found lower ruminal pH values when dried grass was compared with fresh. The lower values of ruminal pH for the grass of Experiment 3A compared with those of Experiment 3B probably reflect the higher water soluble carbohydrate content of the former grass.

The concentration of total volatile fatty acids (TVFA) in the rumen contents when the four diets were fed are shown in Fig. 41. Analysis of variance showed the differences between the dried and fresh grass of Experiment 3B at 09.00 h. was significant. The higher values for the dried grass were associated with a higher intake of dry matter. Williams and Christian (1966) found higher ruminal TVFA concentration with higher intakes of fresh grass. The low TVFA concentration of the fresh compared with the dried grass of Experiment 3A is surprising since dry matter intake and chemical composition of the diets were similar. However, El Shazly (1952) reported higher pre- and post-feeding ruminal TVFA concentrations when fresh grass was compared with frozen grass, and Christian and Williams (1957) reported higher pre-feeding values for a dried grass compared with a fresh grass diet. The higher values for the fresh grass of Experiment 3A (first cut) compared with those of Experiment 3B (third cut) agree with the generally higher values of ruminal TVFA concentrations found by Bath and Rook (1965) for early cuts of rye-grass compared with an autumn regrowth. This is probably due to the higher water soluble carbohydrate content. The high nitrogen content of the late cut grass may also have contributed to the difference since Bryant and Ulyatt (1965) found a reduction in TVFA concentration in the rumen contents of sheep fed grass given a high level of nitrogen fertilization compared with grass given a low level. This effect may, however, have been due to the nitrogenous fertilizer reducing the

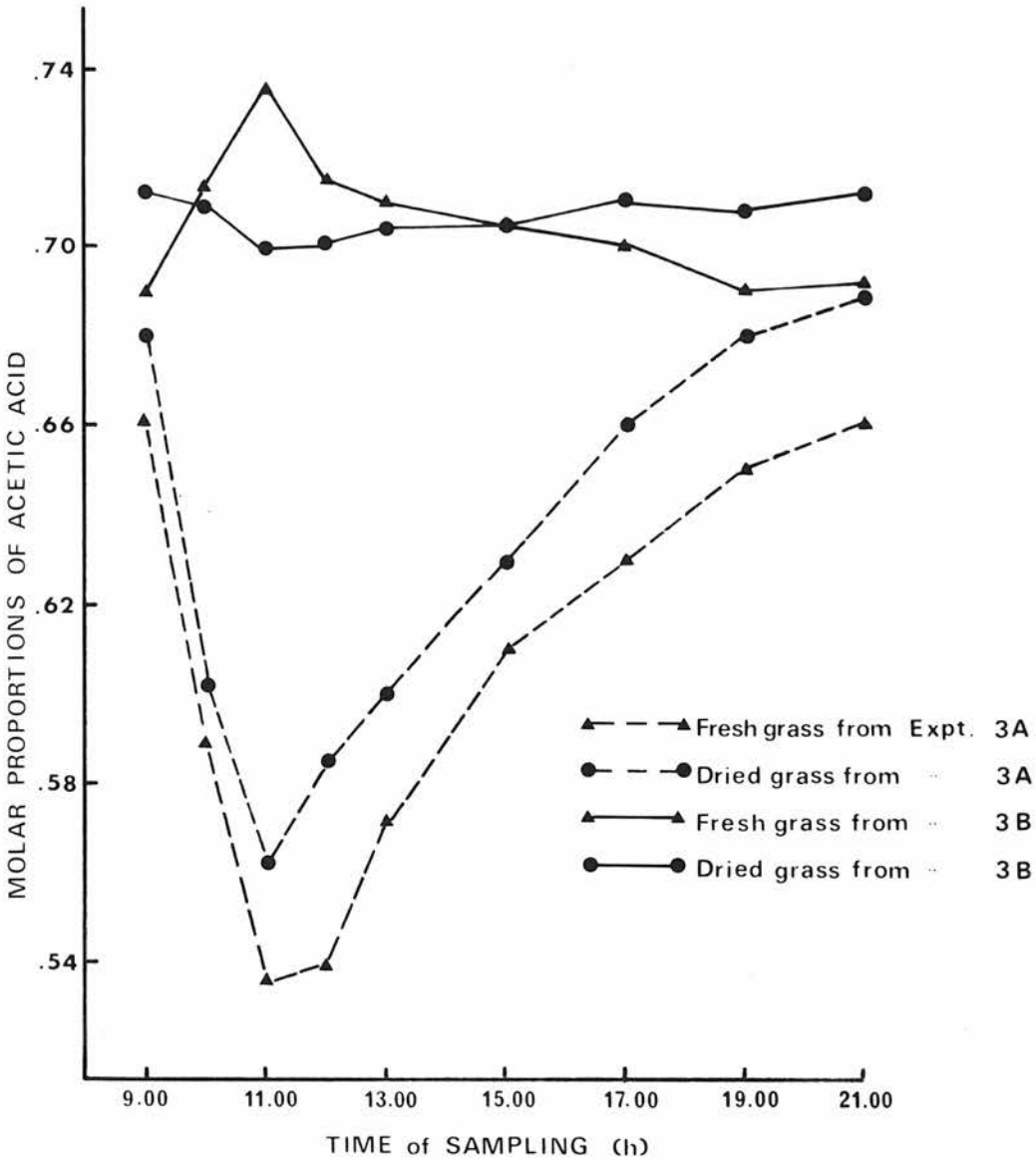

Fig. 42 RUMINAL MOLAR PROPORTIONS OF ACETIC ACID

water soluble carbohydrate content of the grass. The difference in pattern of ruminal TVFA concentration for the dried grass diets in the present investigation, with a later post-feeding time of maximum concentration for the grass of Experiment 3B could reflect the differences in water soluble carbohydrate and fibre contents but differences in dry matter intake may also have contributed.

Major Acids: Fig. 42 shows the molar proportions of acetic acid in the rumen contents when the sheep were fed the four diets. With the fresh and dried grass of Experiment 3A the post-feeding changes in pattern were similar for both diets, although the values were lower for the fresh grass. The post-feeding pattern was not the same for the fresh and dried grass of Experiment 3B; values were lower at pre-feeding but higher immediately after feeding for the fresh grass compared with the dried. Analysis of variance showed a significant difference between the fresh and dried grasses in both experiments at 11.00 h.

El Shazly (1952) found lower pre- and post-feeding values of ruminal acetic acid for frozen grass compared with dried grass. Bryant and Ulyatt (1965) reported an increase in ruminal acetic acid with increased nitrogen fertilization of fresh grass. Thomson and Terry (1965) fed fresh grasses of composition similar to those of Experiments 3A and 3B and reported similar patterns of change in ruminal acetic acid proportions to those in the present investigation. Armstrong (1964) reported mean molar proportions of acetic acid in rumen contents of 0.627 and 0.686 when dried first and third cuts of ryegrass were fed, which is similar to the effect shown here. The higher molar proportions of acetic acid in the rumen contents with the grasses of Experiment 3B compared with those of Experiment 3A reflect the lower water soluble carbohydrate and higher fibre contents of the former diets.

The molar proportions of propionic acid in the rumen contents when sheep

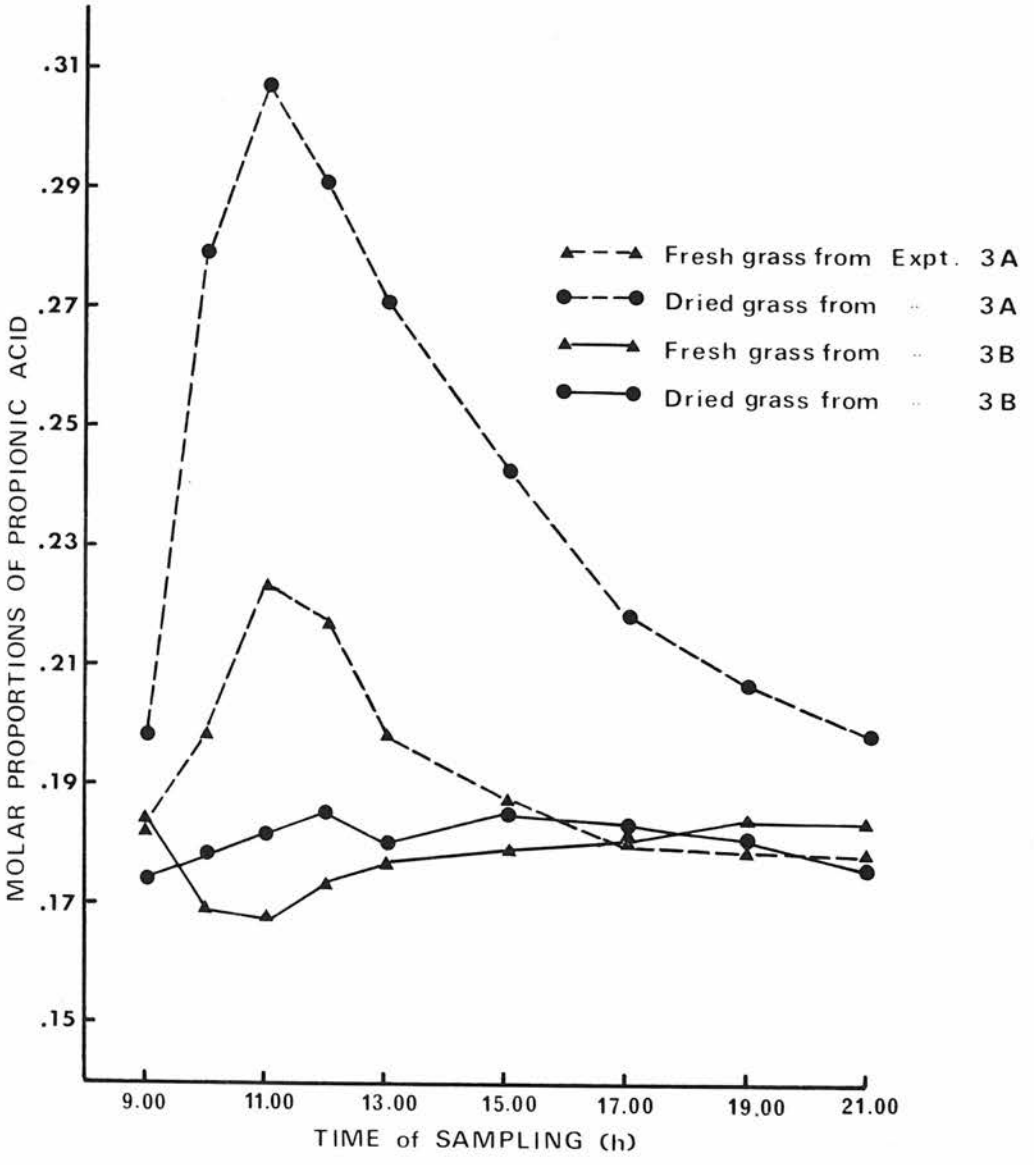

Fig. 43 RUMINAL MOLAR PROPORTIONS OF PROPIONIC ACID

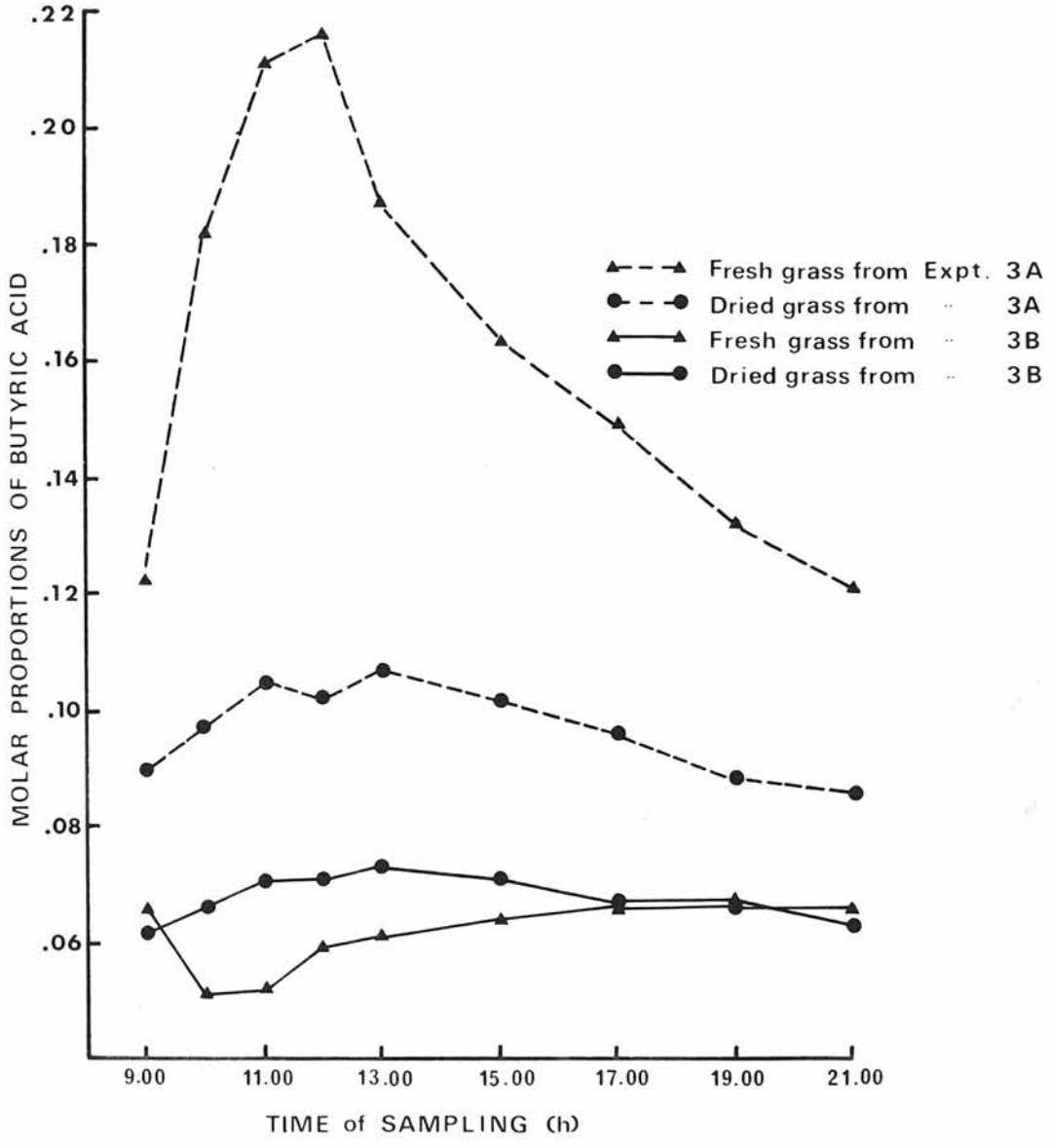

Fig.44 RUMINAL MOLAR PROPORTIONS OF BUTYRIC ACID

were given the four diets are shown in Fig. 43. Analysis of variance gave no significant evidence for differences between the fresh and dried grasses. With the grass of Experiment 3B there was very little difference in pre- and post-feeding values reflecting the changes in acetic acid proportions. For herbages of similar composition Thomson and Terry (1965) reported a similar pattern. While the post-feeding pattern of the proportions of propionic acid in the rumen contents are similar for the fresh and dried grass of Experiment 3A, there are considerable differences in level between the two dietary forms. Thomson and Terry (1965) reported equally high post-feeding values but a narrower range. The post-feeding values of ruminal molar proportions of propionic acid reported by El Shazly (1952) were similar for frozen and dried grass and similar to those for the fresh grass of Experiment 3A.

The relative molar proportions of propionic acid with the fresh and dried grass of Experiment 3A are not in keeping with the associated changes in acetic acid, the dried grass with the higher proportion of acetic acid having the higher propionic acid also. This gives rise to wide differences between the acetic to propionic acid ratios at different stages after feeding. For the fresh and dried grass at 11.00 h. the ratios were 2.4 and 1.9 and at 15.00 h., 2.9 and 2.2 respectively.

Fig. 44 shows the molar proportions of butyric acid in the rumen contents when the four diets were fed. Analysis of variance gave no significant evidence for differences between the fresh and dried grass of Experiment 3B. Differences between the fresh and dried grass of Experiment 3A at 10.00, 11.00, 12.00 and 15.00 h. were significant. The higher values for the fresh grass diet show that in this case lowered ruminal acetic acid levels were reflected by changes in butyric rather than propionic acid as was the case for the dried material. El Shazly (1952) reported pre- and post-feeding molar proportions of butyric acid in

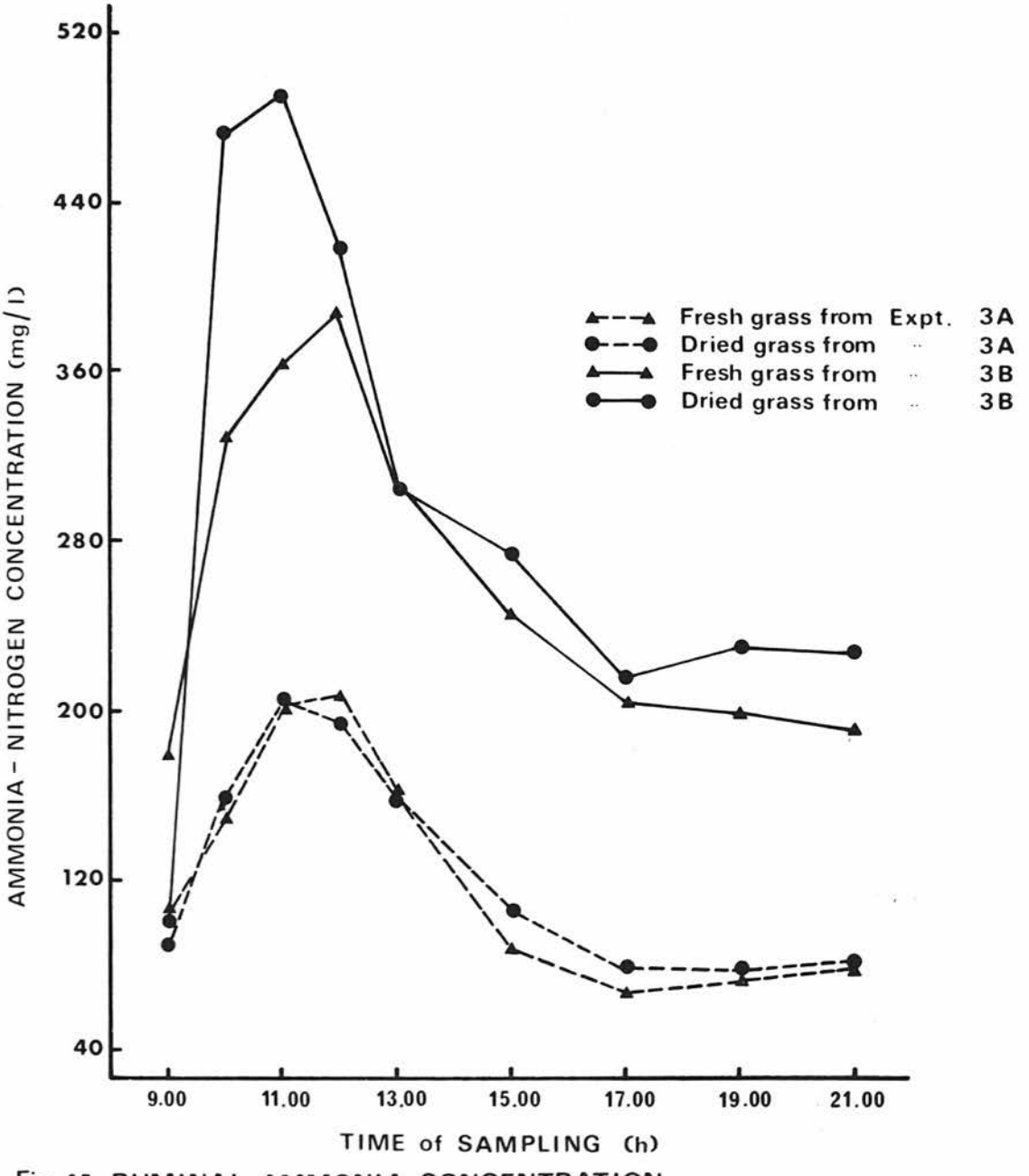

Fig. 45 RUMINAL AMMONIA CONCENTRATION

the rumen contents of sheep fed dried grass similar to those for the dried grass of Experiment 3A, but his values for frozen grass lay between those for the fresh grasses of Experiments 3A and 3B.

Ammonia: Fig. 45 shows the ruminal ammonia concentration curves when the four diets were fed. The grasses of Experiment 3B with the higher nitrogen content showed higher ruminal ammonia concentrations than those for the grass of Experiment 3A. The nitrogen content of the fresh and dried grass of Experiment 3B was the same but intake of the dried grass was higher and this may account for the higher ruminal ammonia concentration with the dried material. Chalmers (1961) when comparing ruminal ammonia curves for diets of frozen and dried grass reported lower values for the latter as did Christian and Williams (1957) for dried compared with fresh grass between two and six hours after feeding. There is no support for the views of the latter workers in the results for the grasses of Experiment 3A where intakes of dry matter were very similar.

CONCLUSION

It is encouraging that the rumen fermentation curves obtained for the dried grass in this experiment confirm those found in the major trials. However, the lack of agreement between the curves for the fresh and dried grasses, particularly for acetic acid in the case of the third cut material, and for propionic and butyric acids for the first cut material, make it dangerous to refine too greatly on differences from fresh grass, based on comparisons using dried grass. The considerable differences in magnitude between the values for fresh and dried materials makes comparisons at various stages invidious.

VII

EXPERIMENT 4.

THE EFFECT ON NUTRITIONAL CHARACTERISTICS OF SILAGES MADE FROM FRESH GRASS, WILTED GRASS AND FORMIC ACID TREATED WILTED GRASS.

INTRODUCTION

Poor animal performance from silage diets can frequently be attributed to the low level of nutrient intake resulting from low voluntary intakes of high moisture silages. Loss of moisture by the wilting of grass prior to ensiling has proved beneficial in increasing the intake of dry matter by sheep, beef and dairy cattle (Murdoch, 1960; Gordon et al., 1961; Harris and Raymond, 1963; Jackson and Anderson, 1968; Alder et al., 1969; Jackson and Forbes, 1970). Adequate consolidation of wilted material in the silo is difficult and trapped pockets of air encourage oxidation of the water soluble carbohydrate of the wilted material causing a rise in temperature. Overheated silages are consumed more readily than unheated materials (Watson and Nash, 1960), but it is generally considered that their nutritive values are reduced.

Application of formic acid to direct cut grass gave restricted oxidation of the water soluble carbohydrate between harvesting and ensiling, and limited the fermentation caused by the Coli-aerogenes bacteria (Henderson and McDonald, 1971; Saue and Breirem, 1969a). Formic acid treated fresh silages resulted in improved animal performance compared with untreated directly ensiled materials (Waldo et al., 1968; 1971; Castle and Watson, 1970a; 1970b; Pike, 1972), and also when compared with wilted silages (Saue and Breirem, 1969b; Waldo et al., 1973). Castle and Watson (1973) noted improved milk yield with formic acid treated wilted silage compared with non-treated wilted silage. Unsupplemented formic acid treated wilted silages have shown improved animal performance compared with untreated wilted silages in trials in the North of Scotland (Swift, 1974), but not in Northern Ireland (Annual Report, 1970), or at Liscombe (MAFF, 1970). The present investigation compares certain nutritional characteristics of fresh and wilted silages, formic acid treated wilted silage and grass, all from the same source of

material. Freezing was used to preserve the fresh cut grass in this trial, since, as shown earlier, rumen fermentation patterns were not the same when fresh and artificially dried grass were fed, and freezing did not affect the digestibility of herbage (Raymond et al., 1953b).

EXPERIMENTAL

The three silages and the grass were fed to eight sheep in a cross-over design, with two sheep on each treatment during one period, as shown in Table 21. Sheep were allocated at random to different treatments and the design was balanced for residual effects.

Table 21

Cross-Over Design for Four Treatments with Eight Sheep

Sheep No / Period	1 (436)	2 (V71)	3 (409)	4 (680)	5 (437)	6 (433)	7 (435)	8 (414)
1	A	B	C	D	A	B	C	D
2	B	C	D	A	B	C	D	A
3	D	A	B	C	D	A	B	C
4	C	D	A	B	C	D	A	B

The treatments were:-

A. Wilted grass silage - 1260 g dry matter fed daily to each sheep.

B. Frozen fresh grass - 789 g dry matter fed daily to each sheep.

C. Formic acid treated wilted silage - 1345 g dry matter fed daily to each sheep.

D. Fresh silage - 863 g dry matter fed daily to each sheep.

The feeding, digestibility and sampling regimes were as described earlier.

The dry matter contents of the diets for the wilted silage, grass, acid treated wilted silage and fresh silage were 316, 175, 336 and 186 g/kg respectively, and the pH values 4.18, 6.10, 4.39 and 3.94. The composition of the diets g/kg dry matter are shown in Table 22.

Table 22

Composition of the Four Diets (g/kg dry matter)

	A (wilted silage)	B (grass)	C (acid treated silage)	D (fresh silage)
Crude protein	142	142	151	144
Crude fibre	287	265	280	298
MAD - fibre	327	289	323	319
Ash	71	70	73	68
Total nitrogen	22.8	22.7	24.2	23.0
Protein nitrogen	6.6	14.7	7.6	5.4
Water soluble nitrogen	16.2	8.0	16.6	17.6
Volatile nitrogen	1.8	0.9	1.6	1.8
Cellulose	286	275	286	302
Acetic acid	24	-	8	36
Propionic acid	0.3	-	0.8	1.7
Butyric acid	0.6	-	0.6	1.4
Lactic acid	59	-	43	102
Succinic acid	nil	-	nil	trace
Water soluble carbohydrate	47	140	151	10
Ethanol	6.4	-	6.1	12

The wilted silage had a reduced total and individual acid content, and increased water soluble carbohydrate content compared with the fresh silage. The silage made by the addition of formic acid to the wilted grass showed an increase in water soluble carbohydrate content and pH, and a reduction in lactic

acid, acetic acid and volatile nitrogen compared with the other two silages.

During the trial, the composition of the grass was investigated after twenty to twenty-four hours removal from cold storage. The water soluble carbohydrate content was reduced, and changes had occurred within the nitrogen fraction. Cores of frozen samples gave a composition identical to the original grass. Since the trial was underway with thawed grass its use was continued and the composition of the grass as fed is shown in Table 22. There were no changes in the water soluble carbohydrate content of high pH, high water soluble carbohydrate, deep frozen silages on defreezing over a period of forty-eight hours.

RESULTS AND DISCUSSION

Nutritive Value

Intake: Mean daily dry matter intakes for the eight sheep were 25.8, 29.6, 32.6 and 22.6 g/kg $W^{0.75}$ for the wilted silage, grass, acid treated wilted silage and the fresh silage respectively. Owing to the restriction placed on the amount of grass provided, this value does not reflect the true intake potential of this material. Average daily intakes for the individual sheep on each treatment are given in Appendix Table 109.

Analysis of variance showed the difference between the intakes of the acid treated wilted silage and the other three materials were significant as was that between the grass and fresh silage. The increase in intake of the wilted silage compared with the fresh silage is in agreement with the increased intake of dry matter with decreased moisture content of the silage reported by many workers (Moore et al., 1960; Murdoch, 1960; 1964; Jackson and Forbes, 1970). Harris and Raymond (1963) reported an increased intake of ryegrass silage, by

sheep, from 17.7 to 49.4 g/kg $W^{0.73}$ when the dry matter of the silage was increased from 153 to 189 g/kg, but wilting to too high a dry matter content (> 400 g/kg) can have an adverse effect on intake (Jackson and Forbes, 1970; McDonald et al., 1968).

Waldo et al. (1968), Nedkvitne (1969), Castle and Watson (1970a, b) and Wilson and Wilkins (1973) have all shown higher intakes of dry matter when unwilted formic acid treated silages were compared with unwilted, untreated materials. Earlier, Wilkins (1970) stated that the addition of formic acid did not result in increased intake compared with untreated silage provided the latter was well preserved. Henderson and McDonald (1971) confirmed this and found no noticeable improvement by formic acid addition. The increase in intake, found in the present work, by the acid treatment of the wilted silage compared with the wilted silage is in agreement with the results from the 1971 trial in the North of Scotland (Swift, 1974), and in Northern Ireland (Annual Report, 1970). They do not agree with the results of the 1972 trial by the North of Scotland College (Swift, 1974), or those at Liscombe (MAFF, 1970). Saue and Breirem (1969b) found no difference between the intakes, by bullocks, of wilted silage and acid treated fresh silage (220 g/kg dry matter).

The intakes of the three silage diets, in the present investigation, are a reflection of their composition. The acid treated wilted silage, with the highest pH and water soluble carbohydrate content, and lower volatile nitrogen content, induced the highest intake of dry matter. As the pH and water soluble carbohydrate content declined so did the intake of silage dry matter. Wilkins (1970) suggested that low intakes of silage dry matter were due to the presence of large quantities of organic acids or extensive degradation of nitrogenous compounds. Later (1971), he stated that intakes of silage dry matter were positively and significantly related to pH. The higher intake of the acid

treated wilted silage confirms these statements.

Digestibility: The mean digestibilities of dry matter, organic matter and nitrogen are given in Table 23, along with estimates of metabolisable energy and gross energy of digestible organic matter. Values for the individual sheep are given in Appendix Table 109.

Table 23

Nutritional Characteristics of the Four Diets

	A (wilted silage)	B (grass)	C (acid treated silage)	D (fresh silage)
Digestibility of dry matter	0.752	0.784	0.776	0.794
Digestibility of organic matter	0.768	0.797	0.788	0.809
Digestibility of nitrogen	0.723	0.752	0.784	0.782
Metabolisable energy (MJ/kg)	11.4	11.6	12.0	13.6
Gross energy of digestible organic matter (MJ/kg)	19.4	19.1	19.8	22.0

As in the previous trials (Experiments 3A and 3B) the digestibility data were adjusted for the level of intake effect by analysis of covariance with intake g/day as the covariate. When adjusted, the digestibility values of organic matter, dry matter, nitrogen and metabolisable energy for the fresh silage, with the lowest intake, were slightly reduced, while for the acid treated wilted silage, with the highest intake, were slightly increased.

Analysis of variance of the data on dry matter and organic matter digestibilities showed the differences between wilted and fresh silage, and between wilted silage and grass, to be significant. Information on the relative

digestibilities of the dry matter of silages made from wilted and unwilted grass are somewhat contradictory. Thus Harris and Raymond (1963) for meadow fescue, Jackson and Forbes (1970), McDonald et al. (1966) confirm the evidence given here of the higher values for unwilted material. While Harris and Raymond (1963) for ryegrass, Hawkins et al. (1970) and Alder et al. (1969) showed the opposite effect.

The use of formic acid in silage making has been claimed to depress dry matter digestibility (Waldo et al., 1971). Henderson and McDonald (1971) showed a decrease in organic matter digestibility for an autumn cut grass ensiled with formic acid compared with untreated material but could not show a similar effect with a first cut grass. Nedkvitne (1969) and Pike (1972) reported increased digestibilities of organic matter as a result of the use of formic acid. The figures presented here indicate that the use of formic acid in the ensiling of wilted material ameliorates the detrimental effect of wilting on digestibility.

Analysis of variance of the digestibility of nitrogen data showed the differences between the wilted silage and the other two silages were significant as was that between the acid treated wilted silage and the fresh grass. McDonald et al. (1966), Durand et al. (1968) and Zelter (1969) have all reported lower digestibilities of nitrogen in prewilted compared with unwilted silages. Waldo et al. (1971) reported similar digestibilities of nitrogen for untreated and formic acid treated silages. Henderson and McDonald (1971) found decreased digestibility of nitrogen for treated compared with untreated silage when a high application of formic acid was added to an autumn cut grass.

Zelter (1969) attributed the higher digestibility of nitrogen for unwilted compared with wilted silage to the higher non-protein nitrogen and ammonia nitrogen of the former. In the present trial, however, the nitrogen fractions

of the wilted and acid treated wilted silages are similar, although the digestibility of nitrogen was much lower with the former diet.

The reduction in the digestibilities of the wilted compared with the other two silages reported here is possibly due to different in-silo temperatures, 31^{o} for the wilted compared with 27^{o} C for the acid treated wilted silage.

Analysis of variance of the metabolisable energy values showed the differences between the fresh silage and the other three diets were significant. Jackson and Forbes (1970) reported slightly higher values of metabolisable energy for unwilted compared with wilted silages, and Jackson (1969) found no difference in metabolisable energy between wilted herbage and silage made from it. Jackson and Anderson (1968) reported metabolisable energy values of 10.5, 10.3 and 10.0 MJ/kg for fresh grass, fresh silage and wilted silage respectively. The metabolisable energy values obtained in the present investigation are to be expected from the effects of the treatments upon digestibility of organic matter and gross energy of digestible organic matter. They confirm the conclusions already reached in Experiments 3A and 3B that there is a positive relationship between metabolisable energy values and the degree of fermentation in the ensiling process.

Rumen Characteristics

The composition of the rumen contents for the individual sheep on each diet are given in Appendix Tables 140 to 141. The values of rumen fermentation products have again been adjusted for the level of intake effect by analysis of covariance, with intake (g/day) as the covariate. The curve patterns have been compared on the basis of first slope, maximum of parabola for time and concentration and the second slope.

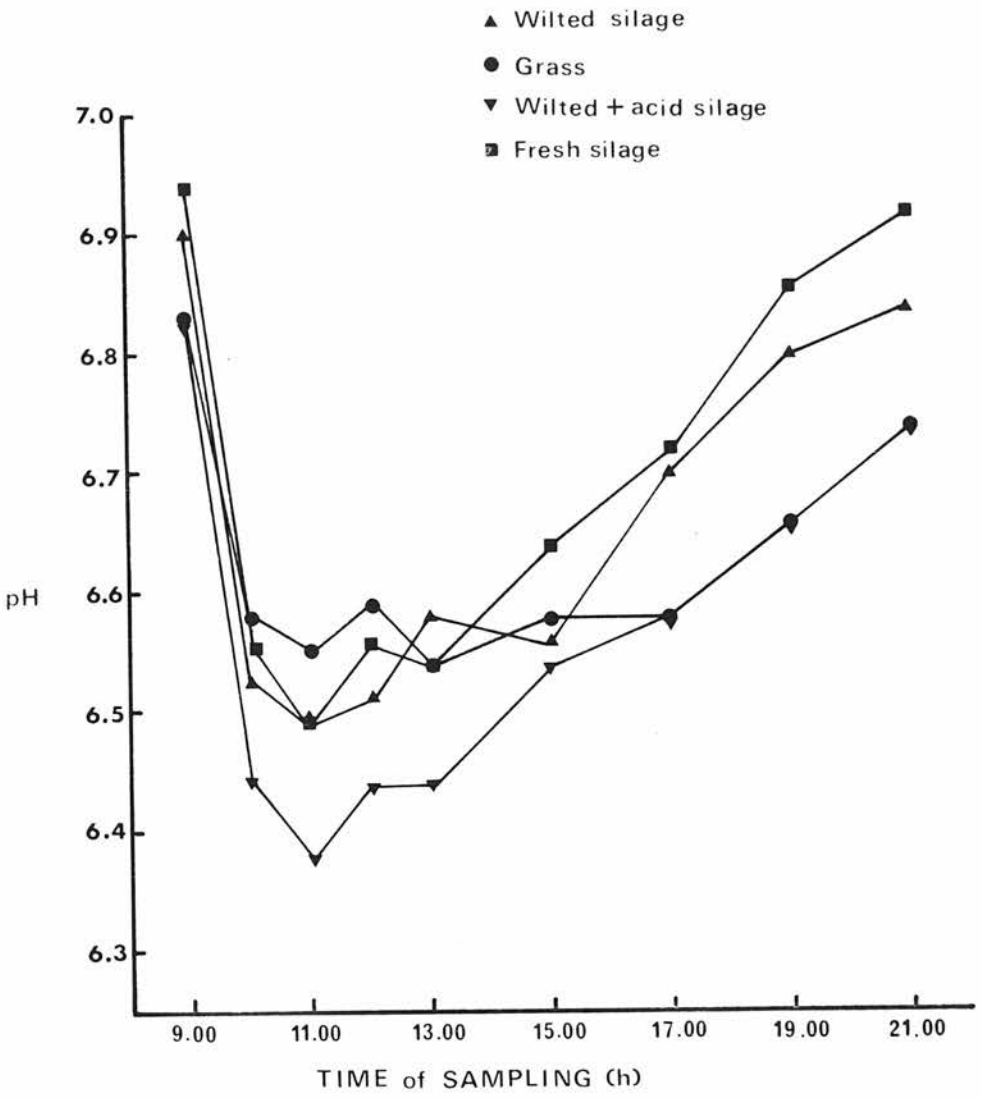

Fig. 46 RUMINAL pH VALUES

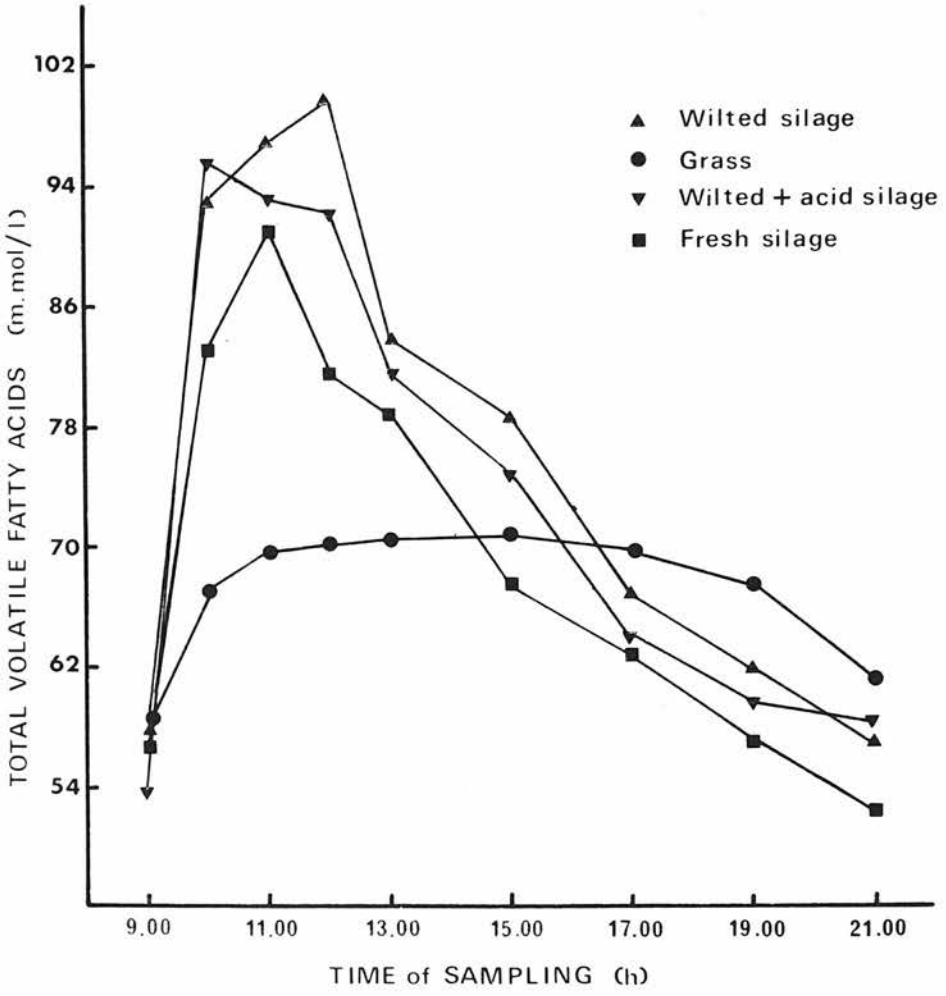

Fig. 47 RUMINAL TOTAL VOLATILE FATTY ACID CONCENTRATION

pH and TVFA concentrations: PH curves of the rumen contents when the four diets were fed are shown in Fig. 46. The curve pattern was similar for all the diets with a decrease following ingestion of food and a gradual rise from the minimum value to the next pre-feeding sample. Analysis of variance gave no significant evidence for differences between the diets at any of the sampling stages, in the time, or concentration, of the minimum of parabola or in the first or second slopes of the curves. However, the times taken to reach the minimum values were different at 2.1, 2.0, 3.3 and 2.6 hours post-feeding for the wilted silage, grass, acid treated silage and fresh silage respectively. Minimum pH values at 6.45, 6.49, 6.31 and 6.47 were little different. The range of values for the different diets is well within those quoted in the literature by Christian and Williams (1957), Terry and Tilley (1964), Du Plessis and Van der Merwe (1970), Balch and Rowland (1957) and Steger et al. (1970) and results obtained by these workers confirm the curve patterns obtained here.

Fig. 47 shows the mean ruminal TVFA concentrations when the four diets were fed. The curve for the grass is different from those for the silage diets. It does not show the same sharp post-feeding rise to a peak value, but a gradual rise to a plateau and consequently there is a narrower range of values for the grass diet. The curve pattern is similar for the three silage diets with a relatively sharp rise after feeding and a gradual fall to the next pre-feeding sample. Maximum values were 102, 98 and 94 m mol/l with the time of maximum of 2.5, 1.7 and 2.3 hours post-feeding for the wilted, acid treated wilted and fresh silages respectively. Analysis of variance confirmed differences between the curves for the grass and the three silages in that the differences between the first and second slopes were significant. The first slope for the fresh silage differed significantly from that for the acid treated material. Surprisingly, differences between the maxima of parabola for time and concentration

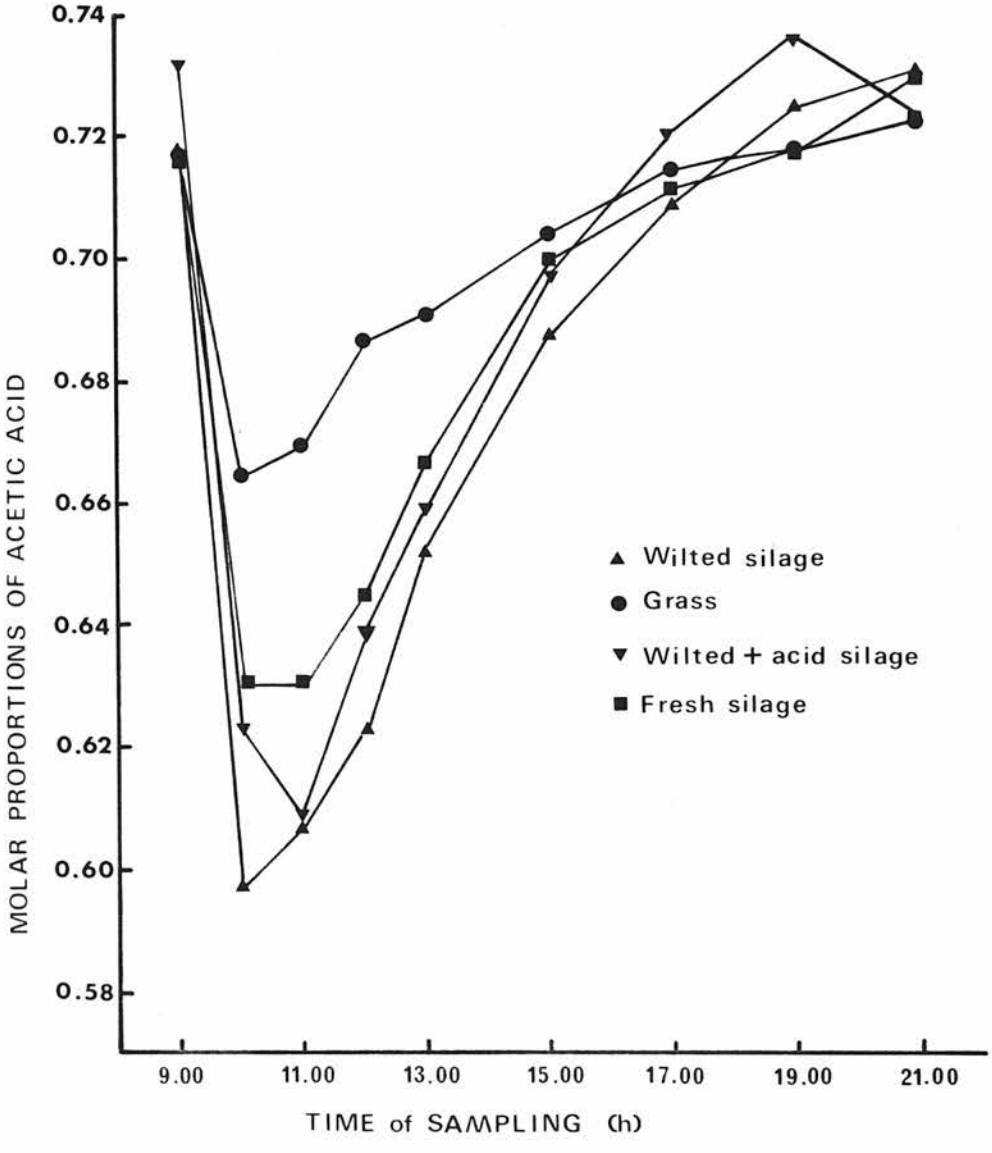

Fig. 48 RUMINAL MOLAR PROPORTIONS OF ACETIC ACID

were not significant. Mean TVFA concentrations for the grass and the wilted silage differed significantly at 10.00, 11.00 and 12.00 h. as did these for the grass and the fresh silage at 11.00 h., and the grass and the acid treated silage at 10.00 and 11.00 h.

El Shazly (1952) reported pre- and post-feeding ruminal TVFA concentrations of 58 and 138 m mol/l for a frozen grass diet. The figures quoted here resemble those of 52 and 81 m mol/l quoted by Christian and Williams (1957) for a fresh grass diet. El Shazly (1952) reported values of 55 to 110 m mol/l for a silage diet. Although the values quoted by Williams and Christian (1959) for ruminal TVFA concentrations with silage diets were lower than in the present work, the patterns of change were similar. Steger et al. (1970) quoted pre-feeding minima of 60 and post-feeding maxima of 90 m mol/l three hours after feeding a maize silage diet. Anderson and Jackson (1971) confirmed lower TVFA concentrations for grass compared with silage diets and, slightly higher values when pre-wilted silage was compared with unwilted material.

The water soluble carbohydrate content of the diet might be expected to be closely related to the post-feeding concentration curves for ruminal TVFA concentration. There is little evidence for this in the present results, particularly as the grass and the acid treated silage which have very different curves have practically the same content of water soluble carbohydrate. The explanation for the differences between the grass and the silages may lie in their different organic acid contents but this does not explain the similarity of the TVFA concentration curves for the silages.

Major Acids: The molar proportions of acetic acid in the rumen contents for the four treatments are shown in Fig. 48. All diets showed the typical post-feeding changes with an initial sharp drop to a minimum proportion followed by a gradual

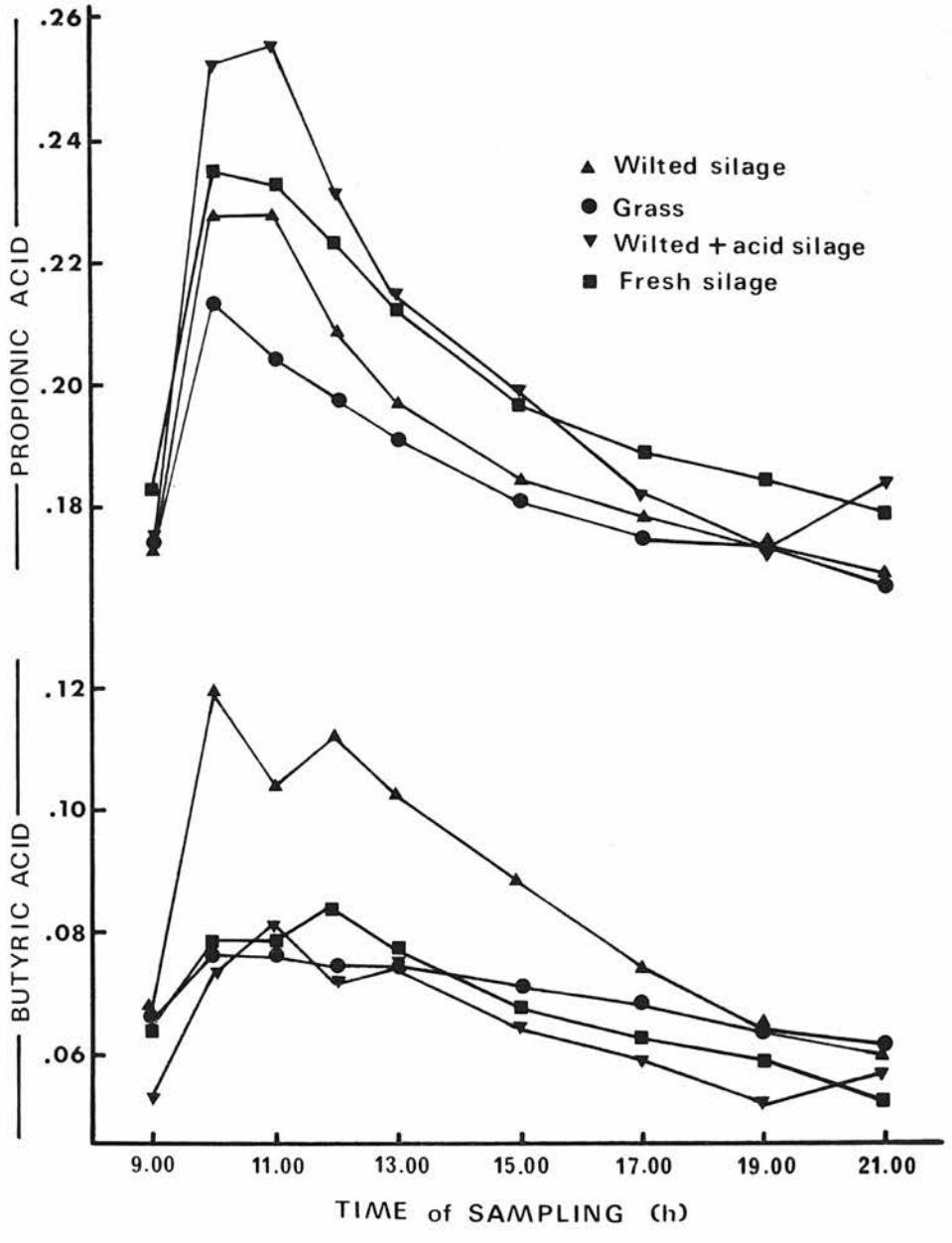

Fig. 49 RUMINAL MOLAR PROPORTIONS OF PROPIONIC AND BUTYRIC ACIDS

rise to the next pre-feeding sample. However, the minimum post-feeding value was higher, and the range of values narrower, for the grass than for the silage diets. Differences between the first slopes of the grass, and the wilted and acid treated silages were significant, as were those between the second slopes of the grass and the silages. The difference between the first slopes of the wilted and fresh silages was significant. The minima of parabola for the grass and the silages differed significantly, as did those for the wilted and fresh silages. Differences between the grass and the other three diets at 10.00, 11.00, 12.00 and 13.00 h., and between the wilted and fresh silages at 10.00 and 13.00 h. were significant.

The molar proportions of propionic acid in the rumen contents when the four diets were given are shown in Fig. 49. The pattern was similar for the four treatments. The rise following ingestion of food and the range of values was less with the grass, reflecting its narrower range of values for acetic acid. There was no significant evidence for differences between the treatments, in the maxima of parabola, for concentration or time, in the first or second slopes, or at any sampling time except 21.00 h., when differences between the wilted and the other two silages and between the grass and the acid treated and fresh silages were significant.

The molar proportions of butyric acid in the rumen contents of sheep fed the four diets are shown in Fig. 49. Analysis of variance gave significant evidence for differences between the first and second slopes for the wilted silage and the other three treatments. Differences between the maxima of parabola, for the wilted silage and both grass and fresh silage, were significant. Differences between the wilted silage and the other three materials were significant at 10.00, 12.00, 13.00 and 15.00 h., as were those between the acid treated silage and the others at 09.00 h.

The values for ruminal acetic acid for the grass diet found here are generally higher than those reported for similar diets by El Shazly (1952), Bath and Rook (1965), Terry and Tilley (1964) and Steger et al. (1970), but follow a similar curve pattern where this is discernible. The levels of acetic acid in the rumen contents of sheep fed the silage diets closely resemble those given by El Shazly (1952) and Bath and Rook (1965) for grass silage diets. Anderson and Jackson (1971) compared wilted with unwilted silage diets and found the molar proportions of acetic acid in the rumen contents was slightly lower for first cut wilted silage, as found here. The results of Puech et al. (1968) confirm the pattern of change in acetic acid proportions obtained in the present investigation, but their wilted and unwilted silages were similar.

The molar proportions of propionic acid in the rumen contents for the grass diet in the present investigation are lower than those quoted by El Shazly (1952), and Terry and Tilley (1964), but this might be expected from the acetic acid levels. The pattern of change following ingestion of food issimilar to that found by Terry and Tilley (1964). The results for the silage diets are higher than those of Bath and Rook (1965) but slightly lower than those of El Shazly (1952). The results for wilted and unwilted silages agree with those of Anderson and Jackson (1971) for first cut silage, and those of Puech et al. (1968). In the present trial the highest maximum proportion of propionic acid in the rumen contents with the silage diets was with the acid treated wilted material which was highest in water soluble carbohydrate. The molar proportions of propionic acid generally reflected those of acetic acid. The acetic to propionic acid ratio throughout the sampling period was generally higher for the grass diet than any of the silages. The ratios for the wilted and acid treated silages were very similar. Those for the fresh silage were slightly lower at all the sampling stages. The ratios for wilted silage were similar to those for lucerne

silages quoted by Zelter (1969), but his ratios for unwilted silage were very much higher, as might be expected.

The molar proportions of butyric acid in the rumen contents, for the grass diet in the present investigation, are considerably lower than those quoted for grass by El Shazly (1952), and Terry and Tilley (1964) but only slightly lower than those of Anderson and Jackson (1971). The results for the silage diets in the present trial are lower than those for similar materials quoted by Bath and Rook (1965) and Puech et al. (1968). Anderson and Jackson (1971) quoted very similar values to those in the present trial of 0.117 and 0.078 for first cut wilted and unwilted silage respectively.

The pattern of change in the molar proportions of ruminal butyric acid for the four treatments was similar, with the narrow range for the grass diet reflecting the acetic and propionic acid values. The large increase following ingestion of the wilted compared with the other two silages is unusual, but reflects the different changes in ruminal acetic and propionic acids at this time.

Orskov et al. (1968) showed that the efficiency of conversion of carbohydrate to volatile fatty acids in the rumen was inversely related to the ratio of butyric to acetic acid in the rumen contents. In the light of this evidence it could be expected that carbohydrate conversion in the rumen on the wilted silage would be lower than for the other diets.

Minor Acids: The molar proportions of iso-butyric, iso- valeric and n-valeric acids in the rumen contents when the four diets were given are shown in Fig. 50. Analysis of variance gave no significant evidence for differences between the diets in the ruminal proportions of iso-butyric acid. However, there were small differences between the diets, the grass having higher pre-feeding values

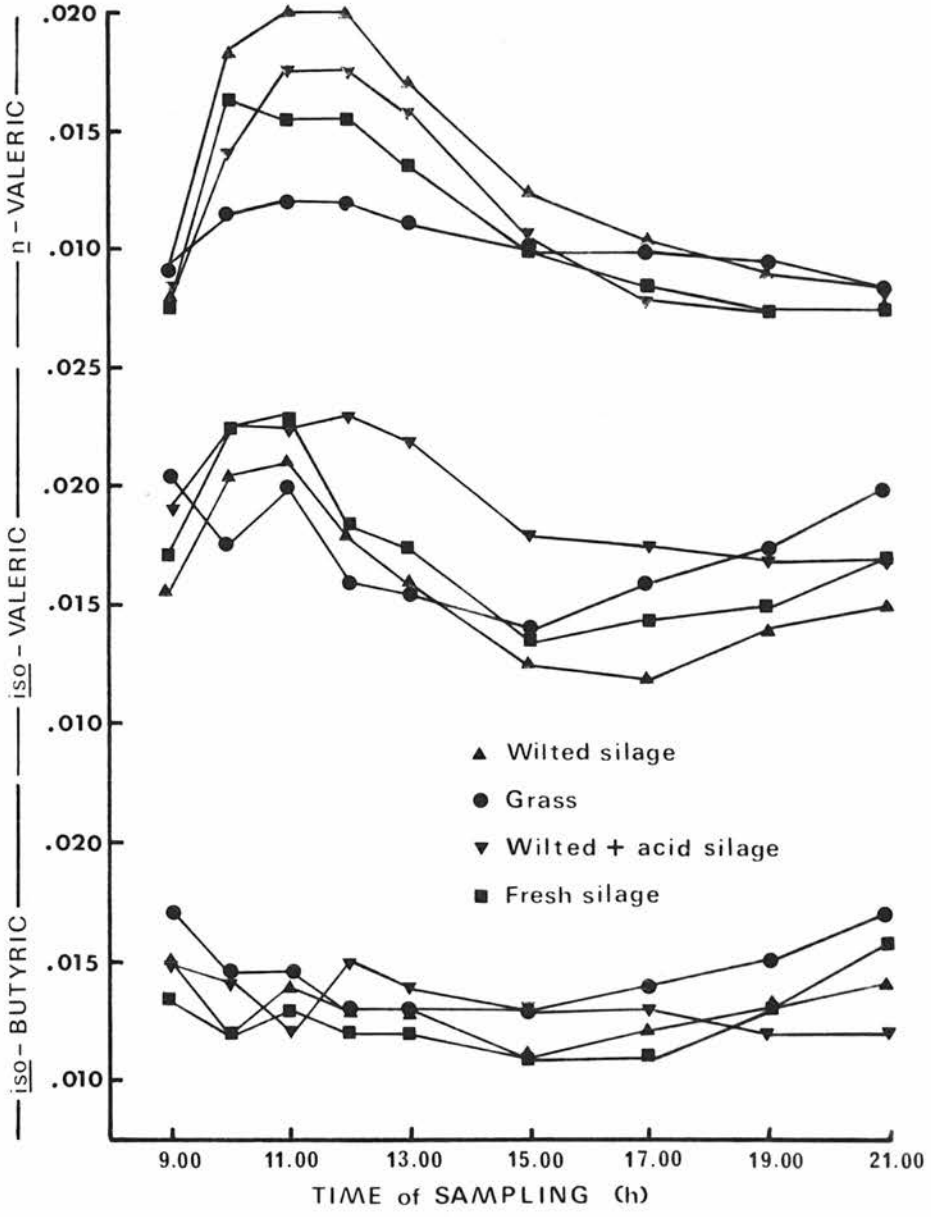

Fig.50 RUMINAL MOLAR PROPORTIONS OF iso - BUTYRIC, iso - VALERIC AND n - VALERIC ACID

compared with the silages. Maximum values were at pre-feeding for all the diets.

Analysis of variance gave no significant evidence for differences between the diets in the ruminal proportions of iso-valeric acid, but there were small differences in the values between the diets. The pattern was different for the grass, with maximum values at pre-feeding, compared with the silages, when maximum values were at two or three hours post-feeding.

The curve pattern of n-valeric acid in the rumen contents was similar for all the diets with minimum values at pre-feeding and maxima at one, two or three hours post-feeding. The maximum values were lowest for the grass and highest with the wilted silage. Differences between the wilted silage and the grass at 10.00, 12.00 and 13.00 h., and between the acid treated silage and the grass at 09.00, 12.00, 13.00 and 19.00 h., and between the fresh silage and the grass at 09.00 and 10.00 h. were significant. As were those between the wilted silage and the acid treated silage at 10.00 h., and between the wilted and fresh silages at 12.00 and 13.00 h.

The curve pattern for ruminal iso-butyric acid proportions with maximum values just before feeding declining after ingestion of food and then rising just before feeding is confirmed by the work of El Shazly (1952), Hawkins et al. (1970) and Schmekel (1967). Fenner et al. (1970), on the other hand, recorded maximum iso-butyric acid proportions one hour after feeding silage. Anderson and Jackson (1971) reported the lower iso-butyric acid proportions for grass compared with silages.

El Shazly (1952) showed a fall in the ruminal proportions of iso-valeric acid, following ingestion of grass, similar to that recorded in the present work. The immediate post-feeding increase in ruminal iso-valeric acid, with the silage diets in the present investigation, agrees with the post-feeding maximum at three hours after feeding a maize silage, reported by Fenner et al. (1970), and the

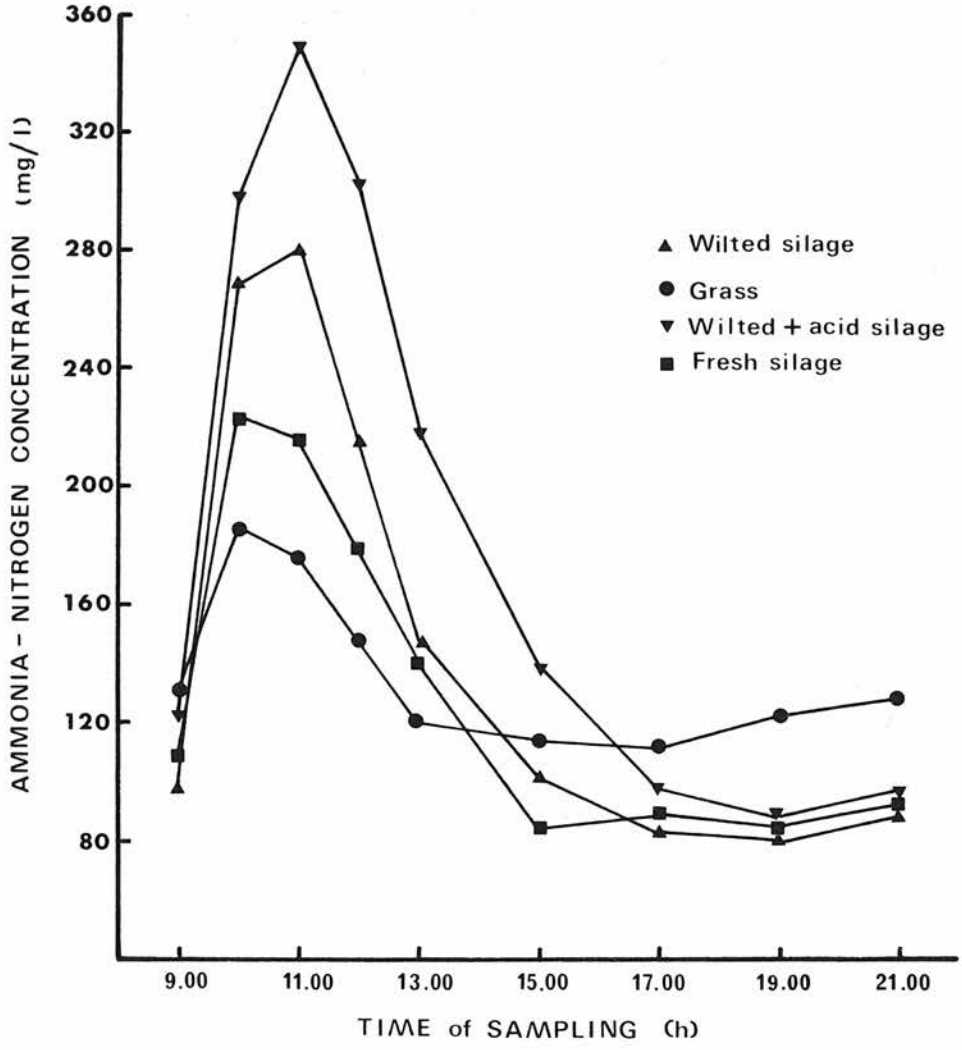

Fig. 51 RUMINAL AMMONIA CONCENTRATION

subsequent drop with these experimental silages to minimum values at six or eight hours post-feeding agrees with the time of minimum values reported by Schmekel (1967) for a silage diet. Hawkins et al. (1970) showed no difference between ruminal iso-valeric acid proportions for wilted and unwilted silages, unlike Anderson and Jackson (1971), who support the slightly higher values for unwilted silage found in the present work.

Since the iso acids are produced by deamination of amino acids, their combined concentration in rumen contents might be expected to reflect the degree of proteolysis in-silo. In the present case the compositional data on volatile nitrogen and soluble nitrogen indicate little or no difference between the silages in the extent of in-silo breakdown of protein, and this is confirmed by the concentrations of the iso acids in rumen contents when the three materials were fed.

The post-feeding increase in ruminal n-valeric acid found in the present investigation confirmed the work of El Shazly (1952) for dried grass and silage diets and of Hawkins et al. (1970) for silages. The curve pattern found here confirms those reported for silage diets by Schmekel (1967). Wilting in this investigation appeared to favour higher levels of ruminal n-valeric acid which is confirmed by the work of Anderson and Jackson (1971), but not by Hawkins et al. (1970).

Ammonia: Ruminal ammonia concentration curves for the four treatments are shown in Fig. 51. The pattern of concentration is similar for all the diets with minimum values at pre-feeding, and maximum value for the grass one hour post-feeding, and for the silages at two hours post-feeding. Analysis of variance showed significant differences between the first slope for the grass and the three silages and for the fresh silage and the other two silages. Differences

between the second slopes for the acid treated silage and the other three diets, and for the grass and the wilted silage were significant. Differences in the maximum of parabola (concentration) between the acid treated silage and the other three treatments, between the wilted silage and the fresh grass, and between the wilted and fresh silages were significant. Differences in the ruminal ammonia concentration between the grass and the wilted silage at 09.00, 10.00, 11.00, 12.00, 17.00, 19.00 and 21.00 h., between the grass and acid treated wilted silage at 10.00, 11.00, 12.00, 13.00, 19.00 and 21.00 h., and between the grass and the fresh silage at 15.00, 17.00, 19.00 and 21.00 h. were significant. Differences between the wilted and acid treated silages at 11.00, 12.00, 13.00 and 15.00 h., between the wilted and fresh silages at 11.00 h., and between the acid treated silage and the fresh silage at 10.00, 11.00, 12.00, 13.00 and 15.00 h. were significant.

Chalmers (1963) reported ruminal ammonia concentration patterns for grass and silage diets similar to those in the present work with higher maximum values for the silage compared with the grass diet. Her minimum values for grass were lower than those for silage, but this was reversed in the present investigation. Hawkins et al. (1970) and Durand et al. (1968) showed ruminal ammonia concentration patterns for wilted and unwilted silages similar to those in the present trial. For silages with a nitrogen content of 27.2 and 27.8 g/kg the former workers reported maximum ruminal ammonia concentrations for wilted and unwilted silages of 303 and 269 mg/l respectively, while Durand et al. (1968) for silages with a higher nitrogen content reported 540 and 420 mg/l respectively. The maximum values for the wilted and unwilted silages, of 283 and 223 mg/l respectively, in the present work support the findings of Hawkins et al. (1970) and Durand et al. (1968). The smaller values in this case reflect the lower nitrogen contents of 22.8 and 23.0 g/kg respectively for the silages.

The maximum ruminal ammonia concentrations achieved on the silage diets in the present investigation are unexpectedly high. The grass curve with its lower post-feeding rise is explicable in terms of the higher water soluble carbohydrate establishing conditions in the rumen conducive to the low ammonia concentrations. A similar effect could be expected with the silages, since the volatile nitrogen and non protein nitrogen did not differ very much between the silages. The order of maximum ruminal ammonia concentration would be expected to be fresh silage, wilted silage and acid treated wilted silage. In fact it is the exact opposite. This is difficult to explain unless the water soluble carbohydrate content is not highly correlated with fermentation activity. There is some evidence for this in the TVFA curves (Fig. 47). The only correlation which could be established between composition of the silages and ruminal ammonia concentration was with the protein nitrogen.

Blood Characteristics

Blood pH, plasma glucose and plasma urea concentrations for the individual sheep are given in Appendix Table 142.

pH: Blood pH values were 7.29, 7.32, 7.32 and 7.28 for the wilted silage, grass, acid treated wilted silage and fresh silage respectively. Analysis of variance gave no significant evidence for differences between the treatments. The minimum value of 7.375 quoted by Vagher et al. (1973), for calves, is slightly higher than the values in the present work, as were the values of 7.39 and 7.47 quoted by L'Estrange and Murphy (1972), for sheep after eighteen days on diets of pelleted grass meal, with and without mineral acids. Ruminal TVFA concentrations, pH values and acetic acid proportions reported by L'Estrange and Murphy (1972) were similar to those for silages in the present investigation. In the present work the addition of formic acid to the silage did not affect the

blood pH. If the pH of the diet affects blood pH, then the blood pH with the grass diet might have been expected to show a higher value, which it did not.

Glucose: Plasma glucose concentrations were 565, 576, 571 and 559 mg/l for wilted silage, grass, acid treated wilted silage and fresh silage respectively. Analysis of variance gave no significant evidence for differences between the treatments. These values are within the range of 550 to 650 mg/l quoted by Reid (1968) for sheep. Bensadoun et al. (1962) found that high plasma glucose levels were associated with narrow ratios of acetic to propionic acid in rumen contents. Bines (1968) could not show higher plasma glucose levels after feeding all concentrate compared with all roughage diets, but did show higher pre-feeding and lower post-feeding plasma glucose concentrations for the all-concentrate diet. His results, however, refer to one cow only. The absence of significant differences among the plasma glucose levels for the three silage diets reflects their similarity of acetic and propionic acid ratios in the rumen contents for these diets. Although there were no significant differences the diets ranked in the same order for plasma glucose concentration as for dietary water soluble carbohydrate content.

Urea: Plasma urea concentrations for the wilted silage, grass, acid treated wilted silage and fresh silage were 173, 152, 158 and 147 mg/l respectively. Analysis of variance gave no significant evidence for differences between the treatments. The higher value for the wilted silage, which had the higher ruminal ammonia concentration, compared with the fresh silage, is in agreement with the results of Durand et al. (1968) and Hawkins et al. (1970) for lucerne silages. The latter workers quoted blood urea concentrations of 156 and 208 mg/l at three hours post-feeding and ruminal ammonia concentrations of 269 and

303 mg/l at one hour post-feeding, for silages of 200 and 400 g/kg dry matter respectively. However, Sutton and Vetter (1971) found no difference in plasma urea concentrations for high and low moisture alfalfa silages, but quoted a higher value for hay compared with silage. The acid treated wilted silage in the present work, despite having the highest concentration of ammonia in the rumen contents did not have the highest plasma urea concentration. This does not agree with the correlation of ruminal ammonia and blood urea found by Abou Akkada and Osman (1967) and Tagari et al. (1964). The acid treated wilted silage did have a higher water soluble carbohydrate content than the other silages and Lewis (1957) reduced blood urea concentration by addition of flaked maize to a diet, but he also reduced ruminal ammonia concentration.

The results of the present investigation tend to confirm the superiority in nutritive value, shown in a number of feeding trials, of silage made from grass treated with formic acid compared with non treated silages. The reduction in digestibility of dry matter and organic matter resulting from wilting are almost completely ameliorated by the addition of acid owing in part to reduction of the in-silo temperature. This results from a reduced oxidation of water soluble carbohydrate which may be expected to lower the dry matter loss. Like the wilted material, but to an even greater extent, fermentation during ensilage of the acid treated material is reduced, compared with untreated, unwilted material. As a result, the gross energy of dry matter is lower and more akin to the original grass. Metabolisable energy values, being largely the resultant of digestibility of organic matter and gross energy are in the order fresh silage, acid treated wilted silage, grass and wilted silage. However, because of differences in dry matter intake, the intakes of metabolisable energy by a 50 kg sheep can be calculated as 5.8, 7.3, 6.5 and 5.6 MJ/day respectively. There is thus a considerable advantage in production potential to the acid treated material and the grass. This probably accounts for the apparent superiority of these materials over directly made silages found in many feeding trials. The present work does not offer an explanation for the similar findings with wilted material.

Ruminal TVFA concentration patterns showed a marked advantage to the fresh grass compared with the silages. The former diet appeared to make available a lower level of acids for a longer period, thus providing a more regular supply of energy and avoiding the flush of acid after feeding, which was characteristic of the silages. Ruminal acetic acid proportions were higher for the grass than the silages. These higher values being outside the range of 0.55 to 0.65 suggested

by Blaxter (1962) as optimal for milk production. The ruminal acetic acid for the grass was reflected by low propionic acid proportions and as a result the acetic to propionic acid ratio was wider with the grass than the silages. It might therefore be expected that the efficiency of utilization of metabolisable energy for growth and fattening would be lower for the grass than the silages.

Although ruminal acetic acid proportions were similar for all the silages, the acid treated wilted silage had a much increased proportion of propionic acid giving a narrower ratio of acetic to propionic acid, which would tend to give higher efficiency of utilization for growth and fattening. The higher molar proportions of butyric acid with the wilted compared with the other silages could be expected to encourage fat production and ameliorate any tendency for low milk fat content as a result of the raised ruminal propionic acid. Neither total ruminal acids nor propionic acid concentrations appear to be related to blood glucose levels.

Although nitrogen contents of the four materials were very similar, the digestibility of nitrogen of the wilted silage was reduced by 0.029 units compared with the grass, while both the directly made silage and the acid treated wilted material showed improved nitrogen digestibilities of 0.030 and 0.032 units respectively, over the fresh grass. In so far as the nitrogen fraction was similar for all three silages, the wilted material could therefore be expected to be the poorest source of nitrogen among the silages. The lower water soluble nitrogen and the very acceptable digestibility of nitrogen make the grass probably the best overall source of nitrogen to the animal. This is confirmed by the ruminal ammonia curves which show lower post-feeding ammonia concentrations for grass than any of the silages. The curves also show that the high nitrogen digestibility for the acid treated material is counteracted by very high ruminal ammonia concentrations which would probably lead to excess wastage of nitrogen voided as urea.

VIII

IMPLICATIONS

Although the primary aim of this work was an examination of the fate of silage in the gut, several subsidiary points, which deserve comment, arose during the course of the work.

Increased frequency of feeding silage improves its dry matter intake and digestibility, and reduces the range of ruminal total volatile fatty acid concentrations. Thus, the animal is given a continuous supply of readily available nutrients in amounts which it can use efficiently. At the same time the deleterious effects on rumen pH, of large intakes of food, are eliminated. The reduction in the range of the molar proportions of acetic and propionic acids means that their ratio remains wide and this might be expected to reduce the efficiency of utilization of energy for growth and fattening, but perhaps improve it for milk production. Thus, present day practice may be wrong, since intensive beef animals are given free access to food, and dairy cows are given concentrates in one or two large meals. It appears that it should be the other way round. Additionally, Robb and Reid (1972) have shown increased efficiency of utilization of acetic acid for fattening when the acid was infused continuously compared with short rapid infusion of the acid into the rumen.

The feeding of dried, rather than fresh, grass gives increased rumen activity as shown by the concentrations of the rumen TVFA, but the effect of drying can vary with the source material. Thus it appears that the drying of spring grass results in a more propionate oriented fermentation and should lead to increased efficiency of utilization, while with the autumn grass the differences between fresh and dried are much less. The fermentation activity is still increased but the differences in the proportions of the major volatile fatty acids are unlikely to affect the efficiency of utilization of energy.

The difference between the compositions of the rumen contents when fresh autumn and spring grass were fed are very considerable and show much reduced

activity as reflected in TVFA concentration. This is at least partly due to the much increased intake of the spring grass. Differences in acetic to propionic acid ratios are large owing to the post-feeding increase in acetic acid concentration with the autumn grass, which is accompanied by a fall in the propionic acid. Thus at the time of maximum fermentation activity the ratio of acetic to propionic was very much wider for the autumn compared with the spring grass. This may well account, in part, for the better animal performance on spring, compared with autumn, grass. Another striking difference between the two grasses is the much higher ruminal butyric acid with the spring grass which is difficult to reconcile with the low milk fat syndrome associated with spring. Rumen ammonia concentrations are much higher with the autumn grass probably owing to its higher nitrogen content. This high nitrogen, however, is typical of autumn grass and may also contribute to poorer animal performance on this material since efficiency of utilization of metabolisable energy will be reduced.

Despite the higher metabolisable energy values of the silages obtained here and confirmed by other workers, silages frequently give disappointing animal performance in practice. At least part of the poor animal performance on silage is explained by reduced intakes of dry matter as silage. It is evident that where fermentation during silage making is restricted, either by wilting or the use of additives, intakes of dry matter and metabolisable energy are improved, even though digestibilities of organic matter are reduced. A similar effect on intake was achieved by delaying the sealing of silos and this effect was particularly noticeable with silage from autumn cut grass. Although each of these treatments reduced digestibility and gross energy of digestible organic matter, the overall intake of metabolisable energy was better than for the corresponding directly ensiled material. The gross energy of silage dry matter was also reduced by reducing the in-silo fermentation.

In the case of wilting and the use of formic acid, the improved production potential is accompanied by relatively low losses of dry matter unless the ensiling technique leads to oxidation loss in the former and leaching loss in the latter. To use delayed sealing as a means of improving intake is extremely risky. Continuing plant respiration will be encouraged with a consequent reduction in the water soluble carbohydrate available for fermentation. This may lead to inadequate acid development for preservation, and secondary fermentations, leading to excessive wastage of silage, particularly in the case of autumn growths of ryegrass heavily fertilized with nitrogen. Similarly, the delayed sealed technique would be extremely risky with legumes or grass species having low water soluble carbohydrate contents such as cocksfoot and meadow fescue.

The higher metabolisable energy values of the silages compared with the grass from which they are made is due largely to their higher gross energy of digestible organic matter, but the improved digestibility of the organic matter of the directly made silages also plays a part. If the determined metabolisable energy values were used to derive k_f [1.] values, and net energy values for growth and fattening calculated, these would be very high. In view of the poor performance found in practice with silage based diets, these would not appear to be valid estimates of the nutritive value. One is forced, therefore, to pose the question of whether the conventionally derived k_f values apply to such foods as silage. It appears unlikely that they do. Thus, the relationship between k_f and metabolisable energy is based on the relationship

$$k_f = 0.03 + 0.81\ q_m \quad [2.]$$

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1. k_f = efficiency of utilization of metabolisable energy for growth or for fattening.

2. q_m = The metabolisability of gross energy of food dry matter $\frac{\text{ME (MJ/kg)}}{\text{GE (MJ/kg)}}$

The assumption is then made that food dry matter has a gross energy of 18.4 MJ/kg which allows the following equation to be derived:

$$k_f = 0.03 + 0.044 \frac{M}{D}$$

With silage, gross energy is of the order of 20 which gives the equation:

$$k_f = 0.03 + 0.040 \frac{M}{D}$$

for silages.

Silages made with restricted fermentation would have lower gross energy of digestible organic matter and it would obviously be invalid to use silage k_f values for these materials. The choice of values to be used in individual cases presents considerable problems. The implications of this discussion to the future of the ARC Metabolisable Energy System would be considerable as the use of different k_f values for different classes of food would further complicate an already complicated system. That this may in fact have to be done receives more support, from the findings of various workers, that the metabolisable energy of pelleted feeds is utilized with different efficiency compared with the non-pelleted parent material. The metabolisable energy of pelleted dried grass is more efficiently used than that of the long grass (Blaxter and Graham, 1955, 1956; Paladines et al., 1964; Wainman et al., 1972).

Efficiency of utilization of energy by the ruminant animal appears to be largely controlled by the relative proportions of the major volatile fatty acids, particularly acetic and propionic, in the final mix absorbed from the rumen. It is pertinent to consider whether there is evidence that these mixes offer any indication of the need for further reduction to the already reduced k_f suggested. The evidence presented here does not lend support for this since in three out of six cases acetic acid was lower and propionic acid higher than for the source material. A fourth

case showed the low acetic acid but this was balanced by an increase in butyric acid. Judged from the point of view of rumen fermentation pattern there is no reason for a further reduction in k_f over that proposed, certainly not in the case of silages made with restricted fermentation. It is interesting in this context that the silage made from autumn cut grass with delayed sealing, although having a lower acetic acid than the grass, at peak fermentation activity showed a balancing increase in butyric and not propionic acid. Although reference has been made previously to the inadequacy of results obtained with dried grass in representing fresh grass, it is considered that their use in this discussion is justified since previous results show dried grass to give a narrower acetic to propionic acid ratio than fresh, and it might be expected that the differences noted between silage and dried grass would in fact have been accentuated if fresh grass had been used. The work of Orskov and Allen (1966) and more recently of Bull et al. (1970) and Robb and Reid (1972) indicate that efficiency of utilization of metabolisable energy may not depend as much upon the relative proportions of acetic and propionic acids in the end products of rumen fermentation as the work of Armstrong and Blaxter (1957b) suggested. In this case the explanation for poor silage performance cannot lie in differences in rumen products.

Ruminal ammonia changes for silage appear to be related to the total nitrogen content and not to the composition of the nitrogen fraction. Generally, ruminal ammonia levels were similar to or lower than those for corresponding dried grass. Fresh grass diets gave lower levels of ammonia than silage diets.

It is clear that the major determinant of the production potential of silage is the intake achieved. The evidence reported here supports the view that intakes are negatively correlated with the degree of lactic acid fermentation taking place during ensilage. It does not enable the major role in reducing intake to be allocated to any particular silage constituent, although low pH, high lactic acid,

high non protein nitrogen were all implicated and materials having low intakes all had low residual water soluble carbohydrate contents. These, however, were all the results of fermentation activity. It is relevant here to note the anomalous results with autumn cut grass ensiled with delayed sealing.

The work described here confirms the need for restricting fermentation during ensilage if the production potential of grass is to be preserved. This may be done by prewilting or by the use of additives coupled with the use of proper techniques in silage making. It may be that the future of herbage conservation lies with sterilisation and not with the process of ensilage.

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APPENDIX

Table 1

Mean Daily Dry Matter Intakes for Individual Sheep

Daily Intake	Treatment	Sheep No.					
		414	419	420	421	V70	V71
g/kg $W^{0.75}$	Silage Diet	37.2	35.0	37.7	42.6	21.7	37.6
	Complete Diet	49.9	68.3	62.2	58.2	50.9	67.2
g	Silage Diet	565	567	543	684	367	729
	Complete Diet	802	1088	847	956	837	1331

Table 2

Composition of Rumen Contents of Sheep No. 414 on the Silage Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.55	53	22.3	0.680	0.222	0.064	0.034
22.00	6.40	205	70.4	0.639	0.289	0.049	0.023
23.00	6.28	242	59.7	0.613	0.285	0.069	0.032
24.00	6.32	187	50.1	0.600	0.270	0.069	0.061
01.00	6.35	96	33.9	0.662	0.242	0.069	0.026
03.00	6.40	51	27.6	0.656	0.241	0.078	0.025
05.00	6.50	77	35.0	0.697	0.208	0.062	0.032
07.00	6.62	76	32.2	0.700	0.201	0.066	0.032
09.00	6.68	65	48.8	0.699	0.194	0.070	0.034
10.00	5.90	204	55.1	0.627	0.293	0.060	0.019
11.00	6.10	213	81.5	0.597	0.287	0.073	0.033
12.00	6.33	189	102.0	0.609	0.276	0.079	0.035
13.00	6.42	132	66.6	0.664	0.226	0.073	0.035
15.00	6.70	64	60.1	0.670	0.230	0.075	0.025
17.00	6.45	40	40.6	0.684	0.206	0.078	0.031
19.00	6.52	65	53.9	0.699	0.202	0.069	0.030
Day 2							
21.00	6.90	92	54.8	0.678	0.210	0.068	0.043
22.00	6.11	295	133.0	0.612	0.264	0.089	0.034
23.00	6.43	221	65.1	0.632	0.236	0.077	0.054
24.00	6.42	166	60.1	0.640	0.243	0.075	0.042
01.00	6.62	97	40.6	0.660	0.226	0.069	0.044
03.00	6.58	74	45.0	0.673	0.207	0.069	0.043
05.00	6.55	62	36.0	0.670	0.213	0.070	0.047
07.00	6.60	112	35.1	0.674	0.216	0.069	0.040
09.00	6.75	102	52.3	0.699	0.202	0.060	0.036
10.00	6.20	205	113.6	0.594	0.293	0.084	0.028
11.00	6.39	217	77.3	0.573	0.286	0.090	0.050
12.00	6.40	96	96.4	0.624	0.236	0.091	0.048
13.00	6.50	75	83.4	0.670	0.219	0.075	0.036
15.00	6.45	142	66.1	0.679	0.213	0.071	0.035
17.00	6.40	105	38.1	0.703	0.201	0.062	0.032
19.00	6.45	104	31.6	0.725	0.186	0.054	0.036

Table 3

Composition of Rumen Contents of Sheep No. 414 on the Complete Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.55	133	36.7	0.713	0.141	0.113	0.033
22.00	–	–	–	–	–	–	–
23.00	6.25	307	140.0	0.631	0.213	0.123	0.032
24.00	–	–	–	–	–	–	–
01.00	5.85	209	99.2	0.653	0.194	0.117	0.034
03.00	6.00	104	73.7	0.659	0.188	0.121	0.033
05.00	6.35	171	62.1	0.665	0.180	0.120	0.034
07.00	6.55	135	50.4	0.667	0.172	0.120	0.040
09.00	6.60	176	52.1	0.681	0.167	0.113	0.039
10.00	–	–	–	–	–	–	–
11.00	6.30	229	122.1	0.628	0.214	0.122	0.035
12.00	–	–	–	–	–	–	–
13.00	4.90	200	99.7	0.649	0.206	0.111	0.032
15.00	6.39	102	67.6	0.647	0.198	0.116	0.037
17.00	6.72	138	54.7	0.655	0.195	0.113	0.035
19.00	6.60	144	58.0	0.665	0.190	0.110	0.035
Day 2							
21.00	6.75	174	38.4	0.668	0.184	0.111	0.037
22.00	–	–	–	–	–	–	–
23.00	6.25	365	119.4	0.619	0.251	0.102	0.028
24.00	–	–	–	–	–	–	–
01.00	6.00	100	69.8	0.627	0.268	0.079	0.026
03.00	6.40	106	46.5	0.639	0.249	0.084	0.027
05.00	6.60	136	48.3	0.665	0.188	0.112	0.034
07.00	6.70	131	41.3	0.670	0.182	0.113	0.033
09.00	6.80	123	29.8	0.674	0.187	0.099	0.040
10.00	–	–	–	–	–	–	–
11.00	5.68	285	130.3	0.611	0.299	0.075	0.015
12.00	–	–	–	–	–	–	–
13.00	5.75	135	111.2	0.625	0.292	0.066	0.017
15.00	6.35	106	92.0	0.660	0.232	0.076	0.031
17.00	6.65	128	44.1	0.689	0.203	0.080	0.028
19.00	6.70	136	32.3	0.706	0.179	0.079	0.036

Table 4

Composition of Rumen Contents of Sheep No. 419 on the Silage Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.75	133	22.7	0.679	0.204	0.074	0.042
22.00	–	–	–	–	–	–	–
23.00	6.65	199	79.8	0.586	0.281	0.076	0.057
24.00	–	–	–	–	–	–	–
01.00	6.75	116	43.0	0.630	0.260	0.073	0.037
03.00	6.80	96	28.7	0.680	0.225	0.064	0.029
05.00	6.90	81	40.9	0.681	0.213	0.061	0.044
07.00	6.85	76	35.0	0.713	0.187	0.066	0.034
09.00	6.90	64	28.1	0.716	0.185	0.063	0.037
10.00	–	–	–	–	–	–	–
11.00	6.70	211	60.0	0.625	0.266	0.066	0.041
12.00	–	–	–	–	–	–	–
13.00	6.58	73	80.0	0.650	0.222	0.067	0.061
15.00	6.68	68	59.0	0.697	0.207	0.069	0.027
17.00	6.87	73	37.2	0.710	0.194	0.067	0.028
19.00	6.80	91	24.0	0.705	0.201	0.063	0.030
Day 2							
21.00	6.82	75	23.4	0.730	0.174	0.064	0.031
22.00	–	–	–	–	–	–	–
23.00	6.45	241	54.8	0.661	0.244	0.056	0.039
24.00	–	–	–	–	–	–	–
01.00	6.65	149	48.7	0.667	0.238	0.065	0.030
03.00	6.85	98	38.0	0.673	0.235	0.065	0.027
05.00	6.70	98	35.8	0.707	0.208	0.060	0.025
07.00	6.85	72	29.9	0.732	0.191	0.052	0.025
09.00	6.80	46	22.3	0.746	0.182	0.046	0.026
10.00	–	–	–	–	–	–	–
11.00	6.30	195	55.0	0.642	0.263	0.058	0.037
12.00	–	–	–	–	–	–	–
13.00	6.48	93	26.8	0.698	0.221	0.053	0.028
15.00	6.58	65	33.0	0.722	0.207	0.054	0.016
17.00	6.50	64	31.0	0.713	0.205	0.059	0.020
19.00	6.70	56	33.6	0.734	0.190	0.051	0.024

Table 5

Composition of Rumen Contents of Sheep No. 419 on the Complete Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.53	122	29.0	0.755	0.150	0.074	0.020
22.00	6.29	403	90.2	0.588	0.237	0.161	0.014
23.00	5.45	242	100.0	0.598	0.258	0.129	0.015
24.00	5.55	196	119.3	0.592	0.278	0.112	0.017
01.00	5.60	201	68.8	0.592	0.272	0.118	0.018
03.00	6.10	187	69.3	0.600	0.251	0.127	0.021
05.00	6.32	116	72.3	0.607	0.219	0.140	0.034
07.00	6.47	114	41.2	0.638	0.214	0.120	0.028
09.00	6.49	106	45.8	0.724	0.155	0.090	0.030
10.00	6.15	474	97.6	0.580	0.237	0.170	0.013
11.00	5.54	368	165.9	0.580	0.260	0.136	0.024
12.00	5.08	300	201.4	0.581	0.268	0.127	0.024
13.00	5.13	201	196.2	0.583	0.277	0.123	0.017
15.00	5.59	153	148.0	0.591	0.266	0.131	0.012
17.00	6.42	120	45.0	0.631	0.232	0.115	0.022
19.00	6.50	106	43.4	0.629	0.216	0.115	0.040
Day 2							
21.00	6.50	92	49.2	0.661	0.208	0.106	0.024
22.00	5.98	520	126.0	0.589	0.224	0.171	0.015
23.00	5.30	306	162.9	0.562	0.273	0.147	0.018
24.00	5.18	300	136.7	0.580	0.258	0.142	0.019
01.00	5.55	201	140.1	0.565	0.275	0.142	0.018
03.00	5.83	169	83.0	0.645	0.223	0.112	0.019
05.00	6.23	147	69.9	0.665	0.215	0.107	0.013
07.00	6.40	96	56.9	0.684	0.198	0.098	0.018
09.00	6.60	85	40.8	0.675	0.198	0.102	0.025
10.00	6.23	418	110.0	0.590	0.220	0.162	0.028
11.00	5.64	350	139.3	0.579	0.259	0.148	0.014
12.00	5.40	201	81.7	0.573	0.280	0.132	0.015
13.00	5.70	127	82.0	0.575	0.290	0.119	0.015
15.00	6.01	150	81.7	0.654	0.231	0.098	0.016
17.00	6.35	105	71.5	0.681	0.205	0.099	0.015
19.00	6.52	104	45.1	0.693	0.190	0.096	0.021

Table 6

Composition of Rumen Contents of Sheep No. 420 on the Silage Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.65	83	37.4	0.691	0.220	0.052	0.036
22.00	–	–	–	–	–	–	–
23.00	6.20	290	81.5	0.598	0.316	0.053	0.032
24.00	–	–	–	–	–	–	–
01.00	6.32	207	55.6	0.608	0.287	0.065	0.038
03.00	6.50	104	56.5	0.669	0.255	0.052	0.023
05.00	6.65	98	60.5	0.678	0.243	0.048	0.030
07.00	6.65	85	53.9	0.699	0.227	0.042	0.033
09.00	6.70	88	36.5	0.708	0.204	0.050	0.038
10.00	–	–	–	–	–	–	–
11.00	6.40	255	57.5	0.585	0.328	0.047	0.039
12.00	–	–	–	–	–	–	–
13.00	6.70	106	50.0	0.660	0.250	0.045	0.045
15.00	6.60	68	44.8	0.690	0.245	0.044	0.020
17.00	6.80	81	50.7	0.699	0.223	0.049	0.028
19.00	6.80	91	35.3	0.714	0.214	0.044	0.027
Day 2							
21.00	6.85	100	38.2	0.720	0.207	0.040	0.033
22.00	–	–	–	–	–	–	–
23.00	6.40	183	58.4	0.641	0.284	0.046	0.028
24.00	–	–	–	–	–	–	–
01.00	6.60	149	44.3	0.645	0.284	0.047	0.023
03.00	6.60	69	37.4	0.688	0.249	0.041	0.021
05.00	6.60	61	31.7	0.690	0.246	0.035	0.028
07.00	6.83	96	35.0	0.694	0.236	0.038	0.032
09.00	6.88	103	36.7	0.700	0.220	0.045	0.041
10.00	–	–	–	–	–	–	–
11.00	6.35	252	82.7	0.588	0.344	0.042	0.026
12.00	–	–	–	–	–	–	–
13.00	6.25	169	53.5	0.636	0.277	0.058	0.027
15.00	6.58	49	44.7	0.677	0.241	0.056	0.025
17.00	6.70	72	44.9	0.679	0.216	0.077	0.027
19.00	6.75	48	24.3	0.707	0.207	0.055	0.030

Table 7

Composition of Rumen Contents of Sheep No. 420 on the Complete Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.50	160	46.2	0.654	0.177	0.118	0.050
22.00	6.21	624	129.8	0.595	0.305	0.080	0.020
23.00	5.45	484	164.1	0.600	0.312	0.074	0.014
24.00	5.23	349	202.2	0.606	0.310	0.069	0.015
01.00	5.35	241	95.2	0.615	0.302	0.068	0.015
03.00	5.85	247	80.7	0.631	0.281	0.071	0.017
05.00	6.12	147	55.1	0.623	0.256	0.090	0.030
07.00	6.35	183	52.4	0.648	0.226	0.100	0.025
09.00	6.60	98	48.0	0.650	0.223	0.101	0.026
10.00	6.23	556	83.4	0.630	0.280	0.076	0.014
11.00	5.50	442	104.9	0.611	0.290	0.081	0.018
12.00	5,38	236	93.8	0.568	0.336	0.081	0.015
13.00	5.41	209	83.0	0.601	0.323	0.064	0.012
15.00	5.96	161	73.6	0.635	0.274	0.074	0.016
17.00	6.42	136	62.1	0.643	0.255	0.084	0.018
19.00	6.56	90	50.5	0.650	0.232	0.091	0.027
Day 2							
21.00	6.63	100	41.9	0.664	0.218	0.093	0.024
22.00	5.90	555	73.1	0.600	0.270	0.075	0.026
23.00	5.15	408	167.9	0.599	0.320	0.069	0.011
24.00	5.40	284	125.0	0.590	0.320	0.067	0.023
01.00	5.53	201	112.0	0.583	0.314	0.084	0.017
03.00	5.90	153	54.8	0.617	0.293	0.077	0.012
05.00	6.30	124	50.0	0.630	0.260	0.090	0.020
07.00	6.40	104	49.3	0.640	0.241	0.097	0.022
09.00	6.65	111	31.1	0.636	0.229	0.105	0.030
10.00	6.24	489	123.1	0.596	0.290	0.094	0.019
11.00	5.60	250	120.7	0.595	0.316	0.075	0.015
12.00	5.18	225	117.6	0.580	0.317	0.086	0.017
13.00	5.40	142	132.6	0.575	0.332	0.079	0.014
15.00	5.95	134	71.0	0.618	0.289	0.077	0.016
17.00	6.50	120	69.3	0.665	0.241	0.078	0.016
19.00	6.55	104	44.9	0.646	0.241	0.089	0.024

Table 8

Composition of Rumen Contents of Sheep No. 421 on the Silage Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.60	99	33.7	0.748	0.178	0.054	0.020
22.00	6.20	213	60.4	0.625	0.288	0.064	0.023
23.00	6.32	283	86.2	0.588	0.282	0.070	0.058
24.00	6.42	204	86.2	0.665	0.239	0.061	0.034
01.00	6.35	128	92.7	0.662	0.244	0.070	0.023
03.00	6.35	94	47.5	0.684	0.227	0.067	0.021
05.00	6.52	101	29.1	0.727	0.197	0.054	0.020
07.00	6.53	91	33.8	0.744	0.181	0.053	0.021
09.00	6.52	74	21.9	0.745	0.174	0.058	0.023
10.00	5.73	270	40.0	0.607	0.275	0.095	0.023
11.00	5.75	303	44.2	0.607	0.318	0.043	0.031
12.00	6.08	300	64.1	0.619	0.287	0.062	0.030
13.00	5.98	240	72.0	0.605	0.273	0.077	0.046
15.00	6.13	120	79.9	0.677	0.222	0.070	0.029
17.00	6.38	64	50.0	0.724	0.200	0.061	0.016
19.00	6.50	41	53.3	0.707	0.201	0.074	0.017
Day 2							
21.00	6.38	42	32.8	0.737	0.183	0.064	0.015
22.00	6.40	277	53.1	0.618	0.294	0.061	0.025
23.00	6.32	298	65.0	0.545	0.326	0.086	0.040
24.00	6.40	252	61.8	0.599	0.273	0.085	0.040
01.00	6.33	217	54.4	0.580	0.283	0.094	0.040
03.00	6.33	104	49.9	0.654	0.241	0.079	0.026
05.00	6.50	77	50.0	0.690	0.217	0.070	0.024
07.00	6.48	80	24.0	0.725	0.193	0.057	0.023
09.00	6.60	68	34.0	0.730	0.175	0.065	0.030
10.00	6.18	205	55.9	0.596	0.306	0.069	0,028
11.00	6.25	258	64.7	0.605	0.304	0.060	0.029
12.00	6.45	225	67.8	0.628	0.262	0.063	0.044
13.00	6.13	120	73.1	0.649	0.248	0.071	0.030
15.00	6.35	102	45.0	0.654	0.244	0.073	0.027
17.00	6.45	37	49.0	0.700	0.215	0.058	0.025
19.00	6.38	80	30.5	0.700	0.219	0.058	0.021

Table 9

Composition of Rumen Contents of Sheep No. 421 on the Complete Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.65	83	30.3	0.659	0.209	0.093	0.039
22.00	-	-	-	-	-	-	-
23.00	5.60	391	76.8	0.608	0.315	0.066	0.011
24.00	-	-	-	-	-	-	-
01.00	5.78	141	81.9	0.593	0.302	0.089	0.015
03.00	6.20	80	81.4	0.599	0.282	0.099	0.020
05.00	6.65	73	64.7	0.603	0.263	0.103	0.030
07.00	6.65	68	40.2	0.647	0.238	0.086	0.028
09.00	6.82	80	27.7	0.631	0.246	0.087	0.036
10.00	-	-	-	-	-	-	-
11.00	5.98	414	55.4	0.588	0.331	0.066	0.014
12.00	-	-	-	-	-	-	-
13.00	5.75	120	75.0	0.580	0.310	0.071	0.039
15.00	6.08	96	60.0	0.607	0.285	0.089	0.017
17.00	6.58	90	58.5	0.664	0.237	0.079	0.020
19.00	6.70	83	44.6	0.670	0.218	0.080	0.032
Day 2							
21.00	6.85	100	30.7	0.699	0.201	0.075	0.024
22.00	-	-	-	-	-	-	-
23.00	5.90	332	105.0	0.566	0.344	0.077	0.013
24.00	-	-	-	-	-	-	-
01.00	5.73	116	134.3	0.572	0.326	0.087	0.015
03.00	6.25	49	61.4	0.615	0.281	0.088	0.018
05.00	6.50	61	42.5	0.631	0.256	0.085	0.028
07.00	6.75	72	45.3	0.659	0.236	0.077	0.028
09.00	6.90	77	30.6	0.650	0.244	0.077	0.029
10.00	-	-	-	-	-	-	-
11.00	5.80	619	139.9	0.571	0.347	0.074	0.008
12.00	-	-	-	-	-	-	-
13.00	5.52	151	119.5	0.593	0.316	0.082	0.008
15.00	6.00	33	61.7	0.593	0.290	0.098	0.018
17.00	6.27	96	56.9	0.616	0.272	0.094	0.017
19.00	6.50	64	53.5	0.636	0.254	0.086	0.024

Table 10

Composition of Rumen Contents of Sheep No. V70 on the Silage Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.55	66	20.5	0.702	0.213	0.062	0.022
22.00	–	–	–	–	–	–	–
23.00	6.45	207	43.6	0.632	0.257	0.071	0.039
24.00	–	–	–	–	–	–	–
01.00	6.40	141	42.7	0.684	0.230	0.056	0.030
03.00	6.55	112	28.7	0.680	0.224	0.071	0.024
05.00	6.70	147	24.8	0.692	0.216	0.069	0.021
07.00	6.65	85	27.6	0.694	0.216	0.064	0.026
09.00	6.74	120	14.9	0.721	0.195	0.057	0.028
10.00	–	–	–	–	–	–	–
11.00	6.40	150	37.8	0.617	0.297	0.062	0.023
12.00	–	–	–	–	–	–	–
13.00	6.50	90	35.0	0.670	0.255	0.060	0.015
15.00	6.57	68	28.6	0.722	0.211	0.053	0.013
17.00	6.70	81	25.8	0.695	0.214	0.068	0.024
19.00	6.65	76	23.7	0.695	0.213	0.062	0.029
Day 2							
21.00	6.65	91	22.2	0.717	0.198	0.055	0.030
22.00	–	–	–	–	–	–	–
23.00	6.30	216	40.1	0.681	0.237	0.051	0.029
24.00	–	–	–	–	–	–	–
01.00	6.33	149	47.1	0.672	0.240	0.053	0.033
03.00	6.45	114	33.1	0.722	0.204	0.051	0.023
05.00	6.60	91	25.4	0.718	0.202	0.056	0.024
07.00	6.63	112	22.0	0.722	0.197	0.050	0.030
09.00	6.68	100	20.3	0.700	0.208	0.059	0.033
10.00	–	–	–	–	–	–	–
11.00	6.52	130	41.3	0.650	0.244	0.063	0.041
12.00	–	–	–	–	–	–	–
13.00	6.55	118	34.3	0.684	0.244	0.055	0.017
15.00	6.60	106	18.7	0.713	0.213	0.044	0.029
17.00	6.58	104	18.5	0.709	0.219	0.043	0.029
19.00	6.65	112	18.7	0.720	0.200	0.050	0.029

Table 11

Composition of Rumen Contents of Sheep No. V70 on the Complete Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.33	175	46.5	0.680	0.156	0.136	0.026
22.00	6.10	418	101.6	0.654	0.227	0.099	0.020
23.00	5.89	500	120.0	0.660	0.224	0.096	0.019
24.00	5.00	383	127.8	0.660	0.228	0.091	0.019
01.00	5.40	241	124.5	0.662	0.228	0.089	0.020
03.00	5.73	255	78.1	0.674	0.208	0.099	0.019
05.00	6.05	170	55.0	0.691	0.196	0.094	0.017
07.00	6.25	144	43.3	0.709	0.174	0.101	0.015
09.00	6.35	123	50.0	0.712	0.165	0.102	0.020
10.00	6.16	507	62.0	0.662	0.235	0.088	0.015
11.00	5.78	483	93.8	0.657	0.218	0.105	0.020
12.00	5.55	402	128.0	0.618	0.262	0.095	0.023
13.00	5.53	348	125.2	0.643	0.249	0.091	0.016
15.00	5.84	201	86.7	0.670	0.237	0.079	0.014
17.00	6.10	161	61.5	0.670	0.208	0.092	0.028
19.00	6.27	57	64.3	0.671	0.193	0.105	0.029
Day 2							
21.00	6.35	133	65.0	0.661	0.207	0.108	0.023
22.00	6.00	564	106.7	0.642	0.259	0.082	0.017
23.00	5.43	366	126.4	0.608	0.291	0.081	0.018
24.00	5.35	300	136.0	0.593	0.314	0.075	0.017
01.00	5.55	224	110.7	0.635	0.273	0.075	0.015
03.00	5.85	209	85.3	0.626	0.265	0.087	0.021
05.00	6.10	186	70.6	0.640	0.244	0.099	0.016
07.00	6.10	177	45.9	0.650	0.225	0.105	0.019
09.00	6.45	111	55.1	0.690	0.186	0.097	0.026
10.00	6.22	497	99.1	0.627	0.273	0.083	0.017
11.00	5.60	425	130.5	0.614	0.301	0.069	0.015
12.00	5.45	337	98.5	0.603	0.311	0.071	0.015
13.00	5.50	217	86.0	0.620	0.292	0.068	0.020
15.00	5.70	205	79.0	0.649	0.265	0.069	0.016
17.00	6.20	187	77.7	0.687	0.222	0.072	0.019
19.00	6.25	209	69.4	0.695	0.203	0.078	0.023

Table 12

Composition of Rumen Contents of Sheep No. V71 on the Silage Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.42	99	45.8	0.726	0.192	0.064	0.018
22.00	6.20	228	81.0	0.612	0.287	0.073	0.027
23.00	6.18	242	86.0	0.636	0.263	0.071	0.029
24.00	6.19	230	91.4	0.666	0.233	0.070	0.031
01.00	6.20	289	67.1	0.666	0.232	0.071	0.031
03.00	6.63	111	43.6	0.691	0.214	0.069	0.026
05.00	6.70	62	41.0	0.711	0.200	0.069	0.020
07.00	6.83	76	40.3	0.751	0.175	0.051	0.022
09.00	6.48	82	41.1	0.719	0.190	0.064	0.026
10.00	6.15	204	66.4	0.628	0.270	0.075	0.026
11.00	6.03	278	110.5	0.617	0.280	0.079	0.023
12.00	6.30	268	94.3	0.647	0.234	0.078	0.040
13.00	6.23	232	70.0	0.629	0.251	0.086	0.034
15.00	6.39	128	56.3	0.690	0.207	0.078	0.025
17.00	6.83	96	58.2	0.701	0.188	0.082	0.029
19.00	6.82	131	39.6	0.733	0.171	0.073	0.022
Day 2							
21.00	6.85	125	55.2	0.724	0.165	0.076	0.036
22.00	6.63	269	82.4	0.673	0.226	0.069	0.030
23.00	6.53	170	62.8	0.660	0.221	0.083	0.034
24.00	6.50	166	62.0	0.690	0.195	0.075	0.040
01.00	6.48	101	62.1	0.726	0.185	0.067	0.022
03.00	6.60	88	39.9	0.729	0.177	0.072	0.023
05.00	6.80	70	32.9	0.762	0.154	0.061	0.022
07.00	6.70	96	30.8	0.759	0.155	0.061	0.024
09.00	6.95	77	28.8	0.725	0.172	0.069	0.032
10.00	6.59	126	58.9	0.694	0.207	0.066	0.032
11.00	6.40	175	69.9	0.690	0.210	0.066	0.033
12.00	6.48	145	73.0	0.696	0.203	0.066	0.034
13.00	6.10	90	82.8	0.697	0.200	0.068	0.033
15.00	6.35	87	70.4	0.715	0.188	0.070	0.027
17.00	6.40	82	51.0	0.747	0.169	0.057	0.028
19.00	6.58	98	39.1	0.736	0.171	0.055	0.037

Table 13

Composition of Rumen Contents of Sheep No. V71 on the Complete Diet

Time of sampling (h)	pH	NH_3-N mg/l	TVFA m mol/l	Molar proportions			
				C_2	C_3	nC_4	$iC_4+iC_5+nC_5$
Day 1							
21.00	6.55	124	47.3	0.641	0.227	0.099	0.033
22.00	–	–	–	–	–	–	–
23.00	5.70	332	74.2	0.612	0.294	0.078	0.016
24.00	–	–	–	–	–	–	–
01.00	5.50	207	94.1	0.626	0.263	0.093	0.017
03.00	5.75	152	83.2	0.663	0.228	0.090	0.018
05.00	6.20	138	72.5	0.662	0.214	0.101	0.023
07.00	6.30	137	61.0	0.684	0.201	0.091	0.023
09.00	6.68	136	32.1	0.709	0.180	0.086	0.025
10.00	–	–	–	–	–	–	–
11.00	5.32	449	96.4	0.620	0.298	0.069	0.013
12.00	–	–	–	–	–	–	–
13.00	5.65	293	105.0	0.600	0.295	0.083	0.022
15.00	5.65	195	89.1	0.586	0.269	0.123	0.021
17.00	6.05	163	80.2	0.620	0.240	0.117	0.023
19.00	6.20	132	93.5	0.634	0.223	0.114	0.028
Day 2							
21.00	6.32	149	62.8	0.713	0.185	0.086	0.016
22.00	–	–	–	–	–	–	–
23.00	5.50	606	117.7	0.635	0.257	0.094	0.013
24.00	–	–	–	–	–	–	–
01.00	5.25	257	89.7	0.571	0.282	0.128	0.018
03.00	5.75	163	86.4	0.580	0.262	0.135	0.023
05.00	6.00	129	79.5	0.602	0.245	0.122	0.030
07.00	6.35	128	58.6	0.648	0.213	0.112	0.027
09.00	6.65	116	41.9	0.700	0.184	0.095	0.021
10.00	–	–	–	–	–	–	–
11.00	5.70	383	96.0	0.592	0.301	0.097	0.011
12.00	–	–	–	–	–	–	–
13.00	5.20	169	96.0	0.597	0.272	0.112	0.019
15.00	5.85	163	79.0	0.628	0.239	0.116	0.016
17.00	6.10	136	72.5	0.617	0.229	0.128	0.024
19.00	6.38	128	67.8	0.664	0.211	0.100	0.025

Table 14

Nutritive Value of Silage fed in Three Ways

	Treatment	Sheep No.								
		88	409	414	434	435	437	447	449	680
Daily intake g/kgW$^{0.75}$	A	58.1	41.0	40.3	44.4	52.6	57.5	49.5	53.1	36.1
	B	45.7	34.5	33.9	26.3	46.2	47.4	38.1	37.6	35.8
	C	49.4	42.6	35.5	45.5	55.6	50.3	40.3	52.6	39.1
Daily intake g/day	A	1383	1066	931	1153	1057	1391	1214	1268	787
	B	1060	883	776	679	924	1148	937	881	769
	C	1152	1111	805	1179	1084	1237	987	1252	861
Dry matter digestibility	A	0.670	0.745	0.735	0.674	0.697	0.680	0.710	0.720	0.711
	B	0.711	0.738	0.735	0.679	0.708	0.696	0.734	0.706	0.721
	C	0.690	0.748	0.752	0.707	0.685	0.702	0.703	0.708	0.735
Organic matter digestibility	A	0.706	0.780	0.772	0.703	0.726	0.709	0.743	0.750	0.745
	B	0.743	0.767	0.772	0.701	0.734	0.734	0.785	0.755	0.757
	C	0.726	0.781	0.793	0.735	0.707	0.735	0.740	0.738	0.773
Nitrogen digestibility	A	0.608	0.628	0.651	0.592	0.563	0.612	0.621	0.628	0.589
	B	0.619	0.654	0.609	0.629	0.610	0.642	0.664	0.592	0.636
	C	0.624	0.657	0.642	0.650	0.587	0.634	0.640	0.597	0.645
Metabolisable energy MJ/kg	A	10.6	11.7	11.5	10.7	10.8	10.7	11.0	11.5	11.3
	B	11.2	11.4	11.4	10.0	11.1	10.9	11.7	11.3	11.3
	C	11.2	11.6	11.9	11.1	10.7	11.0	11.0	11.3	11.7

Table 14 (Cont'd)

Nutritive Value of Silage fed in Three Ways

	Treat-ment	Sheep No.								
		88	409	414	434	435	437	447	449	680
Gross energy of digestible organic matter MJ/kg	A	19.9	19.8	19.8	19.8	19.7	19.9	19.8	20.1	19.8
	B	19.8	19.8	19.8	19.8	19.9	19.8	20.1	20.1	20.0
	C	19.8	19.6	19.6	19.6	20.1	19.8	19.8	19.9	19.7

A = Sheep fed at 12 hour intervals allowed free access
B = Sheep fed at 12 hour intervals restricted access
C = Sheep fed at 4 hour intervals restricted access

Table 15

Composition of Rumen Contents of Sheep No. 88 on Treatment A[1]

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.68	64.9	61	0.751	0.172	0.009	0.050	0.011	0.006	0.002
10.00	6.08	103.8	148	0.633	0.257	0.006	0,076	0.011	0.012	0.005
11.00	6.14	92.4	162	0.624	0.259	0.006	0.079	0.012	0.013	0.007
12.00	6.36	87.2	157	0.649	0.235	0.007	0.080	0.011	0.013	0.006
13.00	6.33	87.5	95	0.675	0.219	0.006	0.077	0.009	0.011	0.004
15.00	6.51	76.9	49	0.716	0.194	0.005	0.067	0.007	0.009	0.003
17.00	6.62	68.1	56	0.741	0.175	0.006	0.062	0.008	0.008	0.002
19.00	6.71	66.2	71	0.743	0.175	0.007	0.058	0.009	0.007	0.002
21.00	6.89	53.2	76	0.750	0.171	0.008	0.051	0.011	0.006	0.003

Table 16

Composition of Rumen Contents of Sheep No. 88 on Treatment B[2]

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.72	56.0	91	0.738	0.183	0.008	0.054	0.008	0.006	0.003
10.00	6.30	81.0	162	0.571	0.243	0.007	0.150	0.011	0.010	0.007
11.00	6.22	84.7	213	0.563	0.237	0.007	0.160	0.012	0.012	0.009
12.00	6.34	86.9	152	0.596	0.224	0.007	0.143	0.011	0.012	0.008
13.00	6.31	80.6	116	0.634	0.211	0.006	0.124	0.008	0.010	0.006
15.00	6.55	75.7	61	0.685	0.201	0.006	0.092	0.006	0.009	0.003
17.00	6.62	69.7	66	0.706	0.195	0.007	0.077	0.006	0.007	0.002
19.00	6.72	65.2	60	0.726	0.190	0.007	0.062	0.007	0.006	0.002
21.00	6.92	57.4	81	0.739	0.188	0.008	0.051	0.009	0.005	0.001

Table 17

Composition of Rumen Contents of Sheep No. 88 on Treatment C(3)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.53	77.3	107	0.710	0.201	0.006	0.062	0.012	0.007	0.002
10.00	6.27	97.5	131	0.643	0.247	0.006	0.075	0.015	0.010	0.005
11.00	6.40	87.5	113	0.702	0.205	0.006	0.062	0.014	0.009	0.003
12.00	6.61	69.3	114	0.711	0.199	0.008	0.059	0.013	0.008	0.002
13.00	6.59	64.7	83	0.720	0.195	0.007	0.055	0.013	0.007	0.003
14.00	6.40	86.5	163	0.646	0.247	0.007	0.064	0.019	0.013	0.006
15.00	6.59	71.3	135	0.672	0.220	0.007	0.070	0.017	0.011	0.004
16.00	6.75	68.9	138	0.687	0.216	0.008	0.061	0.017	0.009	0.002
17.00	6.61	69.7	101	0.713	0.197	0.008	0.060	0.013	0.007	0.002
18.00	6.35	89.7	138	0.647	0.247	0.005	0.068	0.017	0.012	0.005
19.00	6.62	76.3	110	0.697	0.210	0.006	0.062	0.012	0.009	0.003
20.00	6.55	76.6	103	0.709	0.206	0.005	0.060	0.012	0.008	0.002
21.00	6.61	70.5	84	0.714	0.202	0.005	0.060	0.010	0.007	0.002

(1) 12 hour feeding interval, free access
(2) 12 hour feeding interval, limited access
(3) 4 hour feeding interval, limited access.

Table 18

Composition of Rumen Contents of Sheep No. 409 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.75	59.4	91	0.733	0.179	0.008	0.060	0.013	0.006	0.002
10.00	6.48	67.4	197	0.591	0.280	0.008	0.084	0.019	0.012	0.006
11.00	6.45	80.6	203	0.616	0.263	0.008	0.078	0.018	0.012	0.005
12.00	6.63	89.8	152	0.650	0.241	0.007	0.074	0.015	0.010	0.004
13.00	6.68	76.7	132	0.657	0.235	0.007	0.075	0.015	0.009	0.003
15.00	6.73	74.8	111	0.679	0.221	0.007	0.071	0.012	0.008	0.002
17.00	6.75	70.2	88	0.681	0.217	0.007	0.069	0.013	0.008	0.003
19.00	6.84	68.7	86	0.710	0.199	0.008	0.063	0.012	0.006	0.002
21.00	7.00	68.2	91	0.731	0.184	0.008	0.058	0.013	0.005	0.002

Table 19

Composition of Rumen Contents of Sheep No. 409 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.78	58.9	97	0.753	0.167	0.008	0.054	0.009	0.005	0.002
10.00	6.54	83.6	162	0.650	0.242	0.008	0.069	0.012	0.013	0.006
11.00	6.36	97.7	215	0.644	0.242	0.007	0.072	0.011	0.016	0.009
12.00	6.51	87.5	193	0.666	0.226	0.007	0.073	0.009	0.013	0.007
13.00	6.51	86.0	131	0.685	0.214	0.007	0.070	0.008	0.012	0.004
15.00	6.71	03.9	85	0.720	0.191	0.006	0.065	0.006	0.009	0.003
17.00	6.92	59.7	85	0.741	0.176	0.007	0.062	0.007	0.007	0.002
19.00	6.85	56.8	76	0.746	0.171	0.008	0.058	0.009	0.006	0.002
21.00	6.98	53.8	95	0.754	0.167	0.009	0.053	0.010	0.005	0.002

Table 20

Composition of Rumen Contents of Sheep No. 409 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.82	67.2	116	0.696	0.197	0.009	0.075	0.012	0.008	0.003
10.00	6.67	84.9	153	0.661	0.207	0.009	0.093	0.014	0.011	0.006
11.00	6.74	66.7	115	0.656	0.207	0.009	0.098	0.013	0.011	0.005
12.00	6.75	66.5	111	0.665	0.204	0.010	0.095	0.012	0.010	0.004
13.00	6.81	62.9	97	0.706	0.182	0.010	0.078	0.012	0.008	0.004
14.00	6.62	82.1	129	0.625	0.224	0.008	0.107	0.016	0.013	0.008
15.00	6.78	68.1	110	0.653	0.211	0.008	0.096	0.015	0.012	0.005
16.00	6.66	64.3	103	0.672	0.202	0.009	0.090	0.015	0.009	0.004
17.00	6.79	68.2	93	0.700	0.188	0.009	0.082	0.012	0.007	0.002
18.00	6.50	81.3	103	0.669	0.205	0.008	0.091	0.012	0.010	0.005
19.00	6.77	71.6	92	0.671	0.208	0.009	0.087	0.013	0.009	0.003
20.00	6.92	65.4	86	0.689	0.197	0.009	0.083	0.012	0.008	0.003
21.00	6.76	65.7	70	0.691	0.199	0.007	0.080	0.011	0.009	0.004

Table 21

Composition of Rumen Contents of Sheep No. 414 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.96	53.6	99	0.744	0.161	0.011	0.059	0.016	0.006	0.002
10.00	6.51	92.0	167	0.642	0.232	0.009	0.084	0.018	0.010	0.005
11.00	6.64	80.9	185	0.647	0.218	0.009	0.088	0.020	0.012	0.006
12.00	6.73	74.1	141	0.665	0.207	0.008	0.086	0.018	0.011	0.005
13.00	6.70	67.5	84	0.671	0.202	0.007	0.089	0.016	0.011	0.004
15.00	6.73	57.2	71	0.711	0.183	0.008	0.076	0.013	0.008	0.003
17.00	6.81	63.4	66	0.750	0.164	0.009	0.060	0.011	0.005	0.001
19.00	6.95	50.8	88	0.753	0.159	0.010	0.058	0.014	0.005	0.003
21.00	7.04	53.1	92	0.766	0.150	0.011	0.052	0.016	0.004	0.001

Table 22

Composition of Rumen Contents of Sheep No. 414 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.84	50.7	64	0.752	0.157	0.011	0.062	0.013	0.005	0.001
10.00	6.62	69.2	142	0.571	0.237	0.007	0.158	0.013	0.008	0.006
11.00	6.40	94.0	131	0.595	0.210	0.008	0.152	0.016	0.012	0.009
12.00	6.70	83.4	94	0.629	0.196	0.009	0.138	0.013	0.009	0.006
13.00	6.77	72.1	73	0.659	0.183	0.008	0.127	0.012	0.009	0.003
15.00	6.84	62.3	54	0.710	0.163	0.008	0.099	0.010	0.007	0.003
17.00	7.04	47.8	76	0.748	0.152	0.008	0.075	0.011	0.005	0.002
19.00	7.10	46.1	67	0.755	0.150	0.009	0.068	0.012	0.005	0.002
21.00	7.08	46.2	76	0.752	0.159	0.008	0.062	0.011	0.005	0.002

Table 23

Composition of Rumen Contents of Sheep No. 414 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.95	54.9	81	0.735	0.165	0.010	0.068	0.016	0.006	0.002
10.00	6.73	63.8	107	0.683	0.192	0.011	0.077	0.022	0.009	0.005
11.00	6.82	55.4	88	0.689	0.191	0.010	0.077	0.021	0.009	0.003
12.00	6.86	58.7	91	0.712	0.177	0.010	0.073	0.018	0.008	0.002
13.00	6.95	52.6	76	0.722	0.171	0.012	0.070	0.019	0.006	0.001
14.00	6.94	70.5	142	0.665	0.217	0.008	0.073	0.020	0.010	0.006
15.00	6.88	68.8	96	0.675	0.207	0.009	0.075	0.021	0.010	0.004
16.00	6.90	65.3	101	0.698	0.192	0.007	0.075	0.018	0.008	0.003
17.00	7.00	57.6	91	0.702	0.186	0.010	0.073	0.019	0.008	0.002
18.00	6.83	74.3	152	0.652	0.217	0.009	0.083	0.023	0.010	0.006
19.00	6.93	73.7	91	0.677	0.202	0.008	0.079	0.020	0.009	0.005
20.00	6.95	91.1	81	0.685	0.217	0.009	0.063	0.012	0.010	0.004
21.00	7.05	64.3	91	0.739	0.186	0.008	0.051	0.009	0.005	0.001

Table 24

Composition of Rumen Contents of Sheep No. 434 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.75	56.5	91	0.735	0.173	0.007	0.067	0.011	0.005	0.003
10.00	6.31	73.7	167	0.577	0.235	0.008	0.145	0.013	0.012	0.008
11.00	6.28	82.2	152	0.591	0.222	0.008	0.142	0.014	0.014	0.010
12.00	6.50	83.9	116	0.608	0.217	0.007	0.137	0.012	0.012	0.008
13.00	6.73	79.8	101	0.617	0.214	0.007	0.133	0.010	0.011	0.007
15.00	6.42	78.6	61	0.654	0.203	0.006	0.114	0.008	0.010	0.005
17.00	6.66	76.3	66	0.676	0.195	0.008	0.100	0.009	0.009	0.004
19.00	6.64	70.8	55	0.706	0.185	0.007	0.084	0.008	0.007	0.003
21.00	6.70	63.9	56	0.737	0.173	0.008	0.066	0.008	0.006	0.002

Table 25

Composition of Rumen Contents of Sheep No. 434 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.19	67.5	123	0.694	0.203	0.008	0.071	0.013	0.009	0.002
10.00	5.98	100.2	197	0.603	0.292	0.007	0.071	0.014	0.010	0.004
11.00	5.91	111.2	224	0.583	0.303	0.006	0.078	0.015	0.012	0.005
12.00	6.23	99.7	202	0.627	0.262	0.006	0.074	0.014	0.012	0.004
13.00	6.11	94.9	121	0.636	0.255	0.007	0.074	0.013	0.011	0.004
15.00	6.30	73.3	108	0.669	0.229	0.006	0.072	0.012	0.010	0.003
17.00	6.47	70.1	81	0.695	0.209	0.007	0.068	0.011	0.009	0.002
19.00	6.38	68.9	94	0.693	0.204	0.007	0.070	0.014	0.009	0.003
21.00	6.30	64.5	76	0.696	0.207	0.007	0.064	0.014	0.009	0.003

Table 26

Composition of Rumen Contents of Sheep No. 434 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.50	77.6	116	0.702	0.199	0.006	0.070	0.011	0.009	0.002
10.00	6.46	82.9	134	0.684	0.213	0.008	0.071	0.012	0.008	0.004
11.00	6.35	75.1	92	0.689	0.206	0.008	0.074	0.012	0.009	0.002
12.00	6.37	74.3	89	0.698	0.198	0.007	0.074	0.011	0.009	0.003
13.00	6.50	74.9	71	0.706	0.192	0.008	0.072	0.012	0.008	0.002
14.00	6.30	77.6	95	0.655	0.233	0.006	0.077	0.012	0.012	0.006
15.00	6.38	73.8	88	0.667	0.222	0.007	0.076	0.013	0.010	0.005
16.00	6.51	71.3	77	0.703	0.203	0.007	0.066	0.010	0.008	0.003
17.00	6.60	70.5	102	0.717	0.189	0.006	0.066	0.012	0.007	0.003
18.00	6.48	80.1	86	0.686	0.211	0.006	0.071	0.012	0.009	0.005
19.00	6.42	74.5	120	0.680	0.212	0.009	0.075	0.013	0.009	0.003
20.00	6.53	70.3	69	0.697	0.197	0.008	0.074	0.012	0.009	0.003
21.00	6.55	67.2	88	0.726	0.186	0.006	0.064	0.010	0.007	0.002

Table 27

Composition of Rumen Contents of Sheep No. 435 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.79	67.5	81	0.740	0.163	0.008	0.071	0.010	0.006	0.002
10.00	6.06	102.6	213	0.567	0.273	0.005	0.128	0.012	0.010	0.005
11.00	6.25	98.5	172	0.594	0.251	0.007	0.119	0.012	0.011	0.007
12.00	6.36	88.8	137	0.622	0.229	0.007	0.113	0.012	0.011	0.007
13.00	6.29	98.9	96	0.638	0.223	0.006	0.108	0.010	0.010	0.006
15.00	6.63	81.0	66	0.699	0.186	0.006	0.090	0.008	0.007	0.003
17.00	6.55	88.8	61	0.698	0.189	0.007	0.087	0.008	0.007	0.003
19.00	6.75	72.3	61	0.722	0.174	0.007	0.079	0.009	0.006	0.003
21.00	6.70	72.1	71	0.723	0.172	0.008	0.077	0.011	0.006	0.002

Table 28

Composition of Rumen Contents of Sheep No. 435 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.65	82.5	72	0.702	0.205	0.009	0.065	0.013	0.005	0.002
10.00	6.05	107.8	171	0.578	0.262	0.006	0.128	0.012	0.008	0.005
11.00	6.04	99.9	242	0.578	0.255	0.005	0.130	0.013	0.012	0.007
12.00	6.09	102.6	140	0.602	0.235	0.005	0.127	0.012	0.012	0.007
13.00	5.87	101.1	113	0.628	0.222	0.004	0.119	0.010	0.011	0.007
15.00	6.35	83.3	49	0.691	0.196	0.007	0.088	0.008	0.007	0.003
17.00	6.57	69.9	35	0.711	0.188	0.007	0.077	0.009	0.006	0.002
19.00	6.70	65.4	43	0.728	0.180	0.008	0.068	0.010	0.006	0.003
21.00	6.65	63.8	57	0.737	0.175	0.009	0.061	0.011	0.005	0.002

Table 29

Composition of Rumen Contents of Sheep No. 435 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.54	73.9	44	0.720	0.168	0.006	0.087	0.008	0.007	0.004
10.00	6.11	110.4	72	0.646	0.207	0.006	0.112	0.011	0.011	0.008
11.00	6.47	99.4	95	0.664	0.196	0.006	0.106	0.011	0.010	0.007
12.00	6.48	86.8	68	0.672	0.192	0.006	0.104	0.010	0.010	0.006
13.00	6.55	85.0	63	0.698	0.179	0.007	0.094	0.009	0.009	0.004
14.00	6.05	108.8	105	0.619	0.219	0.005	0.126	0.011	0.011	0.009
15.00	6.12	100.8	90	0.634	0.208	0.006	0.120	0.011	0.012	0.008
16.00	6.47	78.7	44	0.667	0.192	0.007	0.107	0.011	0.010	0.006
17.00	6.49	75.4	54	0.680	0.185	0.007	0.102	0.010	0.010	0.006
18.00	6.05	109.9	82	0.613	0.222	0.006	0.130	0.011	0.011	0.008
19.00	6.22	102.2	94	0.655	0.201	0.006	0.111	0.010	0.011	0.007
20.00	6.40	83.4	55	0.677	0.187	0.006	0.104	0.009	0.010	0.006
21.00	6.44	79.3	41	0.684	0.187	0.006	0.098	0.010	0.009	0.005

Table 30

Composition of Rumen Contents of Sheep No. 437 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.56	69.0	61	0.727	0.189	0.007	0.060	0.010	0.006	0.002
10.00	6.15	110.7	162	0.617	0.276	0.007	0.078	0.012	0.009	0.002
11.00	6.11	108.5	174	0.618	0.268	0.007	0.078	0.013	0.010	0.005
12.00	6.24	96.7	165	0.632	0.258	0.007	0.077	0.012	0.010	0.004
13.00	5.82	117.6	124	0.642	0.248	0.005	0.078	0.011	0.011	0.004
15.00	6.50	69.9	76	0.687	0.219	0.006	0.068	0.010	0.008	0.002
17.00	6.62	72.4	58	0.708	0.204	0.007	0.064	0.009	0.007	0.002
19.00	6.43	75.5	47	0.711	0.200	0.007	0.064	0.010	0.007	0.002
21.00	6.19	79.6	50	0.714	0.197	0.007	0.064	0.010	0.007	0.002

Table 31

Composition of Rumen Contents of Sheep No. 437 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.45	71.4	54	0.718	0.193	0.008	0.061	0.013	0.006	0.002
10.00	6.09	111.7	147	0.632	0.263	0.006	0.072	0.014	0.009	0.004
11.00	5.87	103.4	226	0.619	0.262	0.006	0.079	0.016	0.013	0.005
12.00	6.07	91.1	145	0.624	0.258	0.006	0.080	0.015	0.012	0.005
13.00	6.16	88.9	105	0.632	0.248	0.007	0.083	0.015	0.011	0.004
15.00	6.18	85.5	62	0.681	0.217	0.007	0.072	0.012	0.009	0.003
17.00	6.41	79.3	53	0.695	0.206	0.006	0.070	0.013	0.008	0.002
19.00	6.35	71.0	49	0.711	0.197	0.006	0.065	0.013	0.007	0.002
21.00	6.49	68.8	57	0.733	0.183	0.006	0.059	0.012	0.006	0.002

Table 32

Composition of Rumen Contents of Sheep No. 437 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.56	71.5	91	0.704	0.209	0.009	0.057	0.010	0.009	0.003
10.00	6.23	98.5	126	0.659	0.235	0.007	0.069	0.011	0.011	0.008
11.00	6.23	88.7	111	0.681	0.218	0.008	0.068	0.011	0.011	0.004
12.00	6.28	82.4	81	0.692	0.209	0.008	0.067	0.011	0.010	0.004
13.00	6.33	90.4	66	0.707	0.199	0.008	0.064	0.010	0.010	0.003
14.00	6.45	89.4	121	0.654	0.250	0.008	0.061	0.011	0.013	0.005
15.00	6.35	91.1	101	0.669	0.232	0.008	0.065	0.011	0.012	0.004
16.00	6.40	95.8	101	0.683	0.224	0.008	0.064	0.007	0.011	0.003
17.00	6.34	8.45	71	0.695	0.210	0.009	0.063	0.011	0.010	0.002
18.00	6.18	108.6	172	0.638	0.250	0.008	0.069	0.013	0.014	0.008
19.00	6.35	100.5	87	0.664	0.236	0.009	0.065	0.011	0.012	0.004
20.00	6.45	71.6	122	0.689	0.202	0.009	0.072	0.020	0.007	0.002
21.00	6.47	80.8	91	0.701	0.206	0.009	0.061	0.012	0.009	0.002

Table 33

Composition of Rumen Contents of Sheep No. 447 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.92	66.7	99	0.713	0.190	0.010	0.065	0;015	0.006	0.002
10.00	6.55	70.0	154	0.632	0.268	0.007	0.064	0.015	0.010	0.004
11.00	6.33	87.2	141	0.643	0.258	0.006	0.065	0.013	0.011	0.004
12.00	6.48	87.2	123	0.641	0.255	0.006	0.072	0.012	0.009	0.005
13.00	6.45	80.7	106	0.656	0.247	0.006	0.065	0.012	0.009	0.004
15.00	6.52	80.3	81	0.695	0.219	0.007	0.061	0.009	0.007	0.002
17.00	6.57	66.7	75	0.693	0.214	0.005	0.067	0.011	0.007	0.002
19.00	6.77	66.5	97	0.712	0.198	0.008	0.063	0.012	0.006	0.002
21.00	6.87	54.6	105	0.729	0.180	0.008	0.061	0.014	0.006	0.002

Table 34

Composition of Rumen Contents of Sheep No. 447 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.82	57.6	111	0.753	0.155	0,008	0.065	0.011	0.006	0.003
10.00	6.32	79.4	183	0.650	0.226	0.007	0.087	0.014	0.011	0.005
11.00	6.28	91.5	182	0.659	0.217	0.005	0.087	0.013	0.012	0.006
12.00	6.47	86.7	152	0,670	0.206	0.006	0.089	0.014	0.010	0.006
13.00	6.42	81.0	106	0.692	0.193	0.005	0.086	0.010	0.009	0.004
15.00	6.54	80.9	66	0.714	0.180	0.007	0.079	0.009	0.008	0.003
17.00	6.68	76.2	91	0.737	0.168	0.007	0.071	0.009	0.006	0.002
19.00	6.71	70.4	106	0.743	0.164	0.008	0.067	0.010	0.006	0.003
21.00	6.98	59.0	101	0.753	0.155	0.008	0.065	0.011	0.006	0.003

Table 35

Composition of Rumen Contents of Sheep No. 447 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.60	76.5	88	0.704	0.208	0.006	0.065	0.008	0.007	0.002
10.00	6.44	83.6	103	0.672	0.230	0.007	0.067	0.010	0.009	0.006
11.00	6.69	78.7	98	0.687	0.219	0.006	0.067	0.009	0.008	0.003
12.00	6.64	68.8	81	0.703	0.205	0.007	0.067	0.009	0.007	0.002
13.00	6.75	68.3	85	0.716	0.192	0.007	0.064	0.014	0.006	0.002
14.00	6.47	87.0	105	0.665	0.246	0.005	0.061	0.009	0.010	0.005
15.00	6.60	76.2	107	0.694	0.220	0.006	0.060	0.010	0.008	0.003
16.00	6.55	70.1	114	0.700	0.212	0.006	0.065	0.009	0.007	0.002
17.00	6.78	61.6	76	0.715	0.198	0.008	0.062	0.009	0.006	0.002
18.00	6.32	91.2	110	0.652	0.251	0.008	0.065	0.009	0.010	0.006
19.00	6.60	76.4	76	0.673	0.232	0.007	0.066	0.010	0.009	0.004
20.00	6.65	67.1	92	0.697	0.212	0.006	0.067	0.009	0.008	0.002
21.00	6.72	64.0	85	0.707	0.205	0.007	0.063	0.009	0.007	0.003

Table 36

Composition of Rumen Contents of Sheep No. 449 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.80	54.8	105	0.724	0.182	0.010	0.059	0.015	0.008	0.003
10.00	6.44	76.9	153	0.670	0.228	0.007	0.064	0.014	0.011	0.006
11.00	6.45	81.2	162	0.662	0.230	0.006	0.069	0.014	0.012	0.007
12.00	6.50	80.7	123	0.666	0.217	0.006	0.078	0.014	0.013	0.006
13.00	6.53	76.6	93	0.676	0.213	0.007	0.075	0.012	0.011	0.006
15.00	6.42	74.4	79	0.711	0.193	0.005	0.068	0.009	0.010	0.004
17.00	6.59	68.3	75	0.715	0.192	0.004	0.068	0.010	0.008	0.004
19.00	6.67	60.9	80	0.725	0.182	0.008	0.065	0.010	0.008	0.003
21.00	6.77	61.1	101	0.750	0.166	0.007	0.057	0.011	0.007	0.002

Table 37

Composition of Rumen Contents of Sheep No. 449 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.90	48.7	122	0.723	0.180	0.011	0.062	0.014	0.007	0.003
10.00	6.30	73.2	212	0.608	0.271	0.008	0.079	0.015	0.013	0.007
11.00	6.30	96.4	243	0.620	0.262	0.008	0.077	0.013	0.013	0.007
12.00	6.31	83.1	186	0.635	0.247	0.008	0.079	0.013	0.014	0.006
13.00	6.49	71.0	152	0.659	0.230	0.007	0.077	0.011	0.011	0.004
15.00	6.60	68.3	101	0.687	0.211	0.007	0.074	0.009	0.009	0.003
17.00	6.67	65.4	81	0.713	0.192	0.008	0.069	0.009	0.008	0.002
19.00	6.87	53.0	81	0.715	0.189	0.011	0.065	0.012	0.007	0.002
21.00	6.99	50.1	101	0.714	0.190	0.013	0.060	0.015	0.007	0.002

Table 38

Composition of Rumen Contents of Sheep No. 449 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.75	64.4	97	0.716	0.177	0.007	0.079	0.011	0.008	0.003
10.00	6.56	90.2	126	0.654	0.207	0.006	0.098	0.015	0.012	0.008
11.00	6.73	76.5	90	0.680	0.191	0.006	0.093	0.014	0.011	0.007
12.00	6.75	68.3	81	0.691	0.186	0.008	0.089	0.013	0.009	0.005
13.00	6.86	62.7	84	0.711	0.178	0.007	0.081	0.012	0.008	0.003
14.00	6.52	78.5	97	0.674	0.195	0.006	0.093	0.013	0.011	0.008
15.00	6.64	74.5	111	0.683	0.192	0.007	0.089	0.013	0.011	0.005
16.00	6.75	68.3	79	0.709	0.177	0.006	0.084	0.011	0.009	0.004
17.00	6.81	53.7	81	0.713	0.174	0.007	0.082	0.011	0.009	0.004
18.00	6.40	89.8	119	0.648	0.209	0.007	0.095	0.017	0.014	0.010
19.00	6.57	77.6	86	0.672	0.198	0.007	0.092	0.014	0.012	0.006
20.00	6.65	71.6	82	0.691	0.186	0.006	0.088	0.014	0.011	0.005
21.00	6.79	65.1	89	0.712	0.177	0.006	0.081	0.012	0.009	0.003

Table 39

Composition of Rumen Contents of Sheep No. 680 on Treatment A

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.75	66.3	88	0.740	0.171	0.009	0.063	0.010	0.006	0.002
10.00	6.41	87.5	126	0.656	0.218	0.006	0.087	0.013	0.013	0.008
11.00	6.53	85.3	140	0.665	0.205	0.007	0.094	0.013	0.011	0.006
12.00	6.40	83.2	113	0.676	0.205	0.006	0.087	0.010	0.011	0.006
13.00	6.53	79.3	86	0.695	0.196	0.006	0.082	0.008	0.009	0.004
15.00	6.61	75.6	81	0.718	0.185	0.005	0.074	0.007	0.008	0.003
17.00	6.71	67.2	90	0.711	0.188	0.007	0.076	0.009	0.007	0.003
19.00	6.74	64.6	76	0.730	0.177	0.007	0.069	0.009	0.007	0.002
21.00	6.77	71.3	85	0.738	0.167	0.008	0.062	0.018	0.006	0.002

Table 40

Composition of Rumen Contents of Sheep No. 680 on Treatment B

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.70	64.8	116	0.749	0.168	0.011	0.050	0.014	0.006	0.001
10.00	6.43	75.1	141	0.660	0.228	0.008	0.071	0.015	0.012	0.006
11.00	6.28	75.6	195	0.670	0.216	0.007	0.075	0.014	0.012	0.006
12.00	6.48	73.8	146	0.677	0.214	0.007	0.075	0.011	0.011	0.005
13.00	6.44	73.2	93	0.695	0.206	0.005	0.071	0.010	0.010	0.004
15.00	6.65	66.6	76	0.723	0.192	0.007	0.060	0.010	0.007	0.002
17.00	6.65	57.6	81	0.735	0.187	0.007	0.053	0.010	0.006	0.002
19.00	6.74	50.5	83	0.738	0.184	0.007	0.051	0.012	0.006	0.003
21.00	6.83	52.8	88	0.757	0.172	0.010	0.042	0.013	0.006	0.001

Table 41

Composition of Rumen Contents of Sheep No. 680 on Treatment C

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.53	67.4	91	0.719	0.195	0.009	0.057	0.011	0.008	0.002
10.00	6.37	69.2	162	0.663	0.233	0.008	0.064	0.015	0.012	0.006
11.00	6.43	72.1	111	0.684	0.215	0.008	0.066	0.013	0.010	0.004
12.00	6.52	65.5	101	0.703	0.205	0.007	0.061	0.012	0.009	0.003
13.00	6.73	58.9	101	0.714	0.194	0.009	0.061	0.013	0.007	0.002
14.00	6.42	84.0	142	0.667	0.237	0.007	0.061	0.012	0.011	0.005
15.00	6.62	71.5	126	0.683	0.218	0.009	0.064	0.013	0.010	0.004
16.00	6.76	63.5	131	0.706	0.206	0.008	0.058	0.012	0.008	0.002
17.00	6.66	66.7	107	0.713	0.195	0.009	0.062	0.012	0.007	0.003
18.00	6.35	83.8	152	0.654	0.237	0.008	0.071	0.013	0.011	0.006
19.00	6.68	67.7	112	0.693	0.212	0.009	0.062	0.012	0.009	0.003
20.00	6.75	67.4	122	0.716	0.198	0.007	0.060	0.010	0.007	0.002
21.00	6.78	61.3	107	0.718	0.194	0.010	0.055	0.013	0.007	0.003

Table 42

Nutritive Value of Silages and Dried Grass for Individual Sheep

	Treatment	Sheep No.								
		409	414	434	435	437	447	448	449	680
Daily intake g/kg $W^{0.75}$	A	27.7	28.1	24.1	31.0	24.6	9.1	34.5	16.3	21.8
	B	34.8	38.1	37.4	43.6	33.4	26.7	41.9	39.4	28.7
	C	37.1	42.2	37.7	48.7	39.5	36.8	41.6	28.2	41.9
Daily intake g/day	A	646	566	543	529	540	206	716	359	433
	B	816	778	865	735	748	620	886	625	440
	C	876	858	853	847	881	840	866	865	863
Dry matter digestibility	A	0.784	0.810	0.747	-	0.812	0.760	0.748	0.779	0.763
	B	0.759	0.717	0.739	0.727	0.745	0.760	0.692	0.729	0.740
	C	0.771	0.752	0.741	0.744	0.755	0.729	0.776	0.736	0.749
Organic matter digestibility	A	0.817	0.841	0.777	-	0.844	0.804	0.784	0.820	0.803
	B	0.794	0.754	0.769	0.766	0.779	0.798	0.733	0.769	0.778
	C	0.798	0.775	0.760	0.766	0.789	0.751	0.798	0.760	0.777
Nitrogen digestibility	A	0.737	0.752	0.700	-	0.735	0.674	0.677	0.710	0.707
	B	0.682	0.602	0.677	0.638	0.644	0.659	0.607	0.609	0.637
	C	0.674	0.616	0.655	0.608	0.655	0.626	0.678	0.607	0.643
Metabolisable energy MJ/kg	A	13.4	13.9	12.7	-	13.8	13.0	13.0	13.1	13.0
	B	12.9	12.3	12.8	12.7	12.8	13.0	11.9	12.5	12.3
	C	10.5	10.0	10.1	10.0	10.5	9.9	10.5	9.7	10.4

Table 42 (Cont'd)

Nutritive Value of Silages and Dried Grass for Individual Sheep

	Treat-ment	Sheep No.								
		409	414	434	435	437	447	448	449	680
Gross energy of digestible organic matter MJ/kg	A	22.6	22.6	22.6	-	22.6	22.7	22.8	22.5	22.6
	B	21.5	21.6	21.7	21.7	21.6	21.7	21.6	21.6	21.5
	C	17.2	17.3	17.4	17.3	17.4	17.3	17.5	17.5	17.5

A = Control silage
B = Treated silage
C = Dried grass

Table 43

Composition of Rumen Contents for Sheep No. 409 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.97	52.9	65	0.770	0.145	0.013	0.048	0.019	0.005	0.000
10.00	6.53	89.4	205	0.568	0.224	0.011	0.153	0.029	0.012	0.004
11.00	6.43	96.5	253	0.603	0.203	0.011	0.153	0.017	0.012	0.002
12.00	6.63	94.6	196	0.636	0.170	0.011	0.131	0.034	0.012	0.005
13.00	6.51	91.6	126	0.662	0.183	0.009	0.110	0.022	0.010	0.004
15.00	6.83	71.3	61	0.702	0.147	0.013	0.107	0.022	0.007	0.002
17.00	6.98	61.2	61	0.760	0.145	0.010	0.061	0.018	0.006	0.002
19.00	6.97	61.7	87	0.766	0.150	0.010	0.053	0.016	0.005	0.001
21.00	7.13	50.4	83	0.780	0.145	0.011	0.043	0.016	0.005	0.000

Table 44

Composition of Rumen Contents for Sheep No. 409 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.69	76.3	82	0.762	0.155	0.007	0.060	0.010	0.006	0.001
10.00	6.25	101.7	146	0.660	0.187	0.006	0.119	0.015	0.010	0.002
11.00	6.11	103.1	150	0.661	0.177	0.007	0.120	0.017	0.013	0.005
12.00	6.35	95.5	155	0.684	0.173	0.006	0.108	0.014	0.011	0.003
13.00	6.38	90.3	86	0.715	0.162	0.006	0.094	0.011	0.009	0.003
15.00	6.56	86.6	43	0.745	0.155	0.005	0.078	0.009	0.007	0.002
17.00	6.55	87.9	61	0.764	0.153	0.005	0.063	0.008	0.006	0.002
19.00	6.65	86.0	65	0.761	0.158	0.005	0.060	0.009	0.005	0.001
21.00	6.83	76.3	65	0.760	0.155	0.010	0.060	0.009	0.005	0.001

Table 45

Composition of Rumen Contents for Sheep No. 409 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.72	83.1	124	0.684	0.197	0.010	0.087	0.013	0.007	0.001
10.00	5.42	133.8	174	0.600	0.279	0.006	0.101	0.008	0.006	0.001
11.00	5.84	159.8	233	0.589	0.293	0.005	0.097	0.008	0.007	0.001
12.00	6.01	149.0	237	0.571	0.308	0.005	0.099	0.006	0.009	0.001
13.00	6.02	151.0	249	0.596	0.283	0.005	0.099	0.007	0.010	0.001
15.00	6.13	128.6	152	0.639	0.245	0.005	0.095	0.007	0.008	0.001
17.00	6.38	113.3	93	0.666	0.218	0.006	0.090	0.012	0.008	0.001
19.00	6.57	99.8	93	0.674	0.211	0.008	0.089	0.010	0.008	0.002
21.00	6.72	94.8	100	0.689	0.201	0.009	0.081	0.012	0.006	0.001

Table 46

Composition of Rumen Contents for Sheep No. 414 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.10	48.6	106	0.732	0.167	0.015	0.059	0.020	0.005	0.002
10.00	6.51	99.8	182	0.593	0.267	0.010	0.095	0.021	0.011	0.004
11.00	6.41	106.2	171	0.639	0.217	0.010	0.090	0.026	0.014	0.005
12.00	6.50	110.4	190	0.662	0.198	0.010	0.084	0.026	0.013	0.007
13.00	6.60	91.0	105	0.684	0.184	0.011	0.082	0.024	0.011	0.005
15.00	6.70	78.9	80	0.712	0.174	0.009	0.074	0.018	0.009	0.003
17.00	6.98	64.9	106	0.715	0.169	0.011	0.064	0.033	0.006	0.002
19.00	6.99	62.1	84	0.732	0.168	0.014	0.058	0.017	0.010	0.002
21.00	7.09	50.0	87	0.728	0.172	0.016	0.057	0.020	0.006	0.002

Table 47

Composition of Rumen Contents for Sheep No. 414 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.92	49.0	92	0.755	0.148	0.013	0.062	0.013	0.007	0.002
10.00	6.49	86.1	192	0.598	0.245	0.010	0.114	0.014	0.013	0.005
11.00	6.54	103.6	236	0.633	0.216	0.010	0.108	0.014	0.015	0.004
12.00	6.46	92.3	183	0.661	0.195	0.009	0.098	0.012	0.015	0.010
13.00	6.64	83.7	122	0.704	0.173	0.008	0.088	0.009	0.012	0.006
15.00	6.87	69.2	79	0.740	0.159	0.007	0.075	0.007	0.008	0.004
17.00	6.93	58.1	70	0.756	0.155	0.008	0.067	0.007	0.006	0.002
19.00	7.07	55.6	87	0.755	0.153	0.011	0.064	0.010	0.005	0.002
21.00	7.16	42.5	92	0.755	0.151	0.013	0.060	0.013	0.006	0.001

Table 48

Composition of Rumen Contents for Sheep No. 414 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.90	54.4	116	0.700	0.170	0.012	0.094	0.014	0.008	0.003
10.00	5.84	112.1	159	0.599	0.276	0.005	0.104	0.007	0.008	0.002
11.00	5.94	104.6	146	0.597	0.271	0.005	0.108	0.005	0.012	0.002
12.00	6.34	115.9	112	0.623	0.243	0.004	0.113	0.005	0.011	0.002
13.00	6.52	85.2	65	0.639	0.226	0.004	0.115	0.003	0.011	0.002
15.00	6.47	94.5	39	0.682	0.192	0.004	0.107	0.003	0.009	0.003
17.00	6.66	86.8	43	0.712	0.176	0.005	0.093	0.005	0.008	0.002
19.00	6.83	72.8	69	0.713	0.170	0.008	0.091	0.008	0.008	0.003
21.00	6.95	71.5	103	0.717	0.168	0.010	0.084	0.011	0.008	0.002

Table 49

Composition of Rumen Contents for Sheep No. 434 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.81	43.8	134	0.740	0.161	0.014	0.061	0.016	0.006	0.001
10.00	6.51	74.9	215	0.643	0.189	0.011	0.116	0.015	0.015	0.012
11.00	6.26	81.5	241	0.631	0.192	0.012	0.125	0.014	0.015	0.012
12.00	6.15	81.1	202	0.669	0.180	0.011	0.104	0.012	0.014	0.011
13.00	6.36	77.4	168	0.695	0.172	0.010	0.093	0.011	0.010	0.010
15.00	6.49	76.0	112	0.741	0.158	0.009	0.073	0.008	0.007	0.003
17.00	6.63	62.7	95	0.759	0.155	0.008	0.057	0.010	0.010	0.002
19.00	6.78	56.9	103	0.754	0.152	0.012	0.063	0.012	0.006	0.002
21.00	6.78	51.8	116	0.756	0.152	0.013	0.057	0.014	0.007	0.002

Table 50

Composition of Rumen Contents for Sheep No. 434 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.78	47.0	110	0.717	0.171	0.014	0.070	0.019	0.008	0.002
10.00	6.28	93.0	190	0.605	0.250	0.012	0.103	0.014	0.012	0.005
11.00	6.10	105.5	170	0.590	0.261	0.010	0.108	0.013	0.013	0.006
12.00	6.19	102.2	173	0.621	0.238	0.010	0.096	0.013	0.015	0.007
13.00	6.23	98.3	85	0.645	0.222	0.010	0.091	0.013	0.013	0.007
15.00	6.47	79.4	102	0.681	0.199	0.011	0.080	0.012	0.012	0.005
17.00	6.33	77.7	80	0.700	0.188	0.011	0.074	0.012	0.011	0.005
19.00	6.55	71.4	73	0.718	0.178	0.015	0.065	0.014	0.008	0.003
21.00	6.68	68.8	100	0.728	0.171	0.013	0.061	0.015	0.009	0.002

Table 51

Composition of Rumen Contents for Sheep No. 434 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.72	73.5	61	0.633	0.242	0.011	0.091	0.013	0.008	0.002
10.00	5.42	120.5	175	0.563	0.331	0.006	0.087	0.005	0,006	0.001
11.00	5.84	145.0	249	0.515	0.373	0.004	0.096	0.005	0.007	0.001
12.00	6.01	142.3	240	0.513	0.371	0.003	0.099	0.005	0.009	0.001
13.00	6.02	129.0	187	0.537	0.338	0.004	0.103	0.005	0.011	0.001
15.00	6.13	116.3	105	0.580	0.299	0.005	0.100	0.006	0.010	0.001
17.00	6.38	94.6	66	0.620	0.265	0.007	0.092	0.007	0.008	0.001
19.00	6.57	90.8	57	0.637	0.255	0.008	0.083	0.009	0.007	0.001
21.00	6.72	81.9	75	0.653	0.236	0.010	0.082	0.010	0.008	0.002

Table 52

Composition of Rumen Contents for Sheep No. 435 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.09	43.1	93	0.727	0.180	0.019	0.048	0.020	0.007	0.001
10.00	6.50	97.0	173	0.521	0.328	0.011	0.104	0.020	0.014	0.002
11.00	6.42	99.7	221	0.538	0.302	0.012	0.101	0.021	0.018	0.007
12.00	6.56	98.0	148	0.610	0.248	0.013	0.085	0.020	0.018	0.007
13.00	6.73	79.4	102	0.638	0.228	0.013	0.085	0.018	0.013	0.006
15.00	6.96	60.6	59	0.706	0.192	0.014	0.062	0.016	0.009	0.003
17.00	7.11	59.8	51	0.720	0.183	0.012	0.053	0.023	0.009	0.002
19.00	7.17	55.0	93	0.732	0.181	0.013	0.051	0.016	0.007	0.002
21.00	7.14	49.1	77	0.734	0.181	0.014	0.046	0.016	0.007	0.001

Table 53

Composition of Rumen Contents for Sheep No. 435 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.95	67.6	70	0.735	0.175	0.010	0.064	0.009	0.007	0.002
10.00	6.06	117.4	222	0.570	0.272	0.006	0.130	0.008	0.010	0.004
11.00	6.24	119.3	279	0.565	0.274	0.007	0.129	0.009	0.013	0.004
12.00	6.09	119.6	230	0.614	0.233	0.008	0.114	0.007	0.017	0.006
13.00	6.19	117.4	131	0.666	0.206	0.007	0.097	0.006	0.014	0.005
15.00	6.44	91.7	44	0.709	0.187	0.007	0.080	0.004	0.010	0.003
17.00	6.70	86.3	52	0.726	0.187	0.007	0.068	0.005	0.006	0.002
19.00	6.91	76.0	66	0.741	0.178	0.007	0.060	0.006	0.007	0.001
21.00	6.99	71.6	57	0.746	0.176	0.008	0.056	0.007	0.006.	0.001

Table 54

Composition of Rumen Contents for Sheep No. 435 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC6
09.00	6.62	89.3	86	0.669	0.205	0.009	0.096	0.011	0.009	0.001
10.00	5.50	125.8	142	0.585	0.309	0.004	0.086	0.008	0.008	0.001
11.00	5.48	142.6	202	0.584	0.312	0.004	0.086	0.005	0.008	0.001
12.00	6.00	120.3	202	0.557	0.319	0.006	0.098	0.006	0.013	0.001
13.00	6.18	109.3	155	0.572	0.298	0.007	0,100	0.007	0.016	0.001
15.00	6.17	108.0	69	0.620	0.261	0.004	0.095	0.004	0.011	0.004
17.00	6.44	102.8	39	0.662	0.227	0.005	0.091	0.005	0.009	0.001
19.00	6.73	97.3	65	0.681	0.207	0.007	0.089	0.008	0.008	0.001
21.00	6.86	90.6	56	0.684	0.201	0.009	0.087	0.010	0.008	0.001

Table 55

Composition of Rumen Contents for Sheep No. 437 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.86	54.1	65	0.750	0.157	0.012	0.059	0.016	0.005	0.002
10.00	6.53	73.7	172	0.624	0.205	0.012	0.118	0.023	0.011	0.007
11.00	6.43	72.2	155	0.650	0.182	0.010	0.107	0.028	0.014	0.009
12.00	6.56	73.0	150	0.679	0.174	0.010	0.096	0.024	0.011	0.006
13.00	6.52	77.2	90	0.705	0.165	0.010	0.087	0.021	0.009	0.004
15.00	6.79	65.2	65	0.734	0.160	0.009	0.070	0.017	0.007	0.003
17.00	6.57	70.9	78	0.749	0.155	0.009	0.064	0.016	0.007	0.002
19.00	7.00	58.6	69	0.748	0.157	0.011	0.061	0.016	0.006	0.001
21.00	6.94	58.7	73	0.755	0.154	0.012	0.057	0.016	0.005	0.001

Table 56

Composition of Rumen Contents for Sheep No. 437 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.64	64.6	70	0.765	0.153	0.012	0.048	0.015	0.007	0.002
10.00	6.27	95.4	183	0.627	0.226	0.008	0.106	0.017	0.010	0.005
11.00	6.23	107.9	205	0.631	0.215	0.009	0.108	0.020	0.012	0.006
12.00	6.28	98.5	170	0.662	0.201	0.008	0.097	0.017	0.011	0.005
13.00	6.16	108.2	118	0.694	0.183	0.008	0.090	0.013	0.010	0.003
15.00	6.50	86.8	70	0.716	0.175	0.007	0.081	0.012	0.008	0.002
17.00	6.66	73.0	57	0.723	0.174	0.008	0.077	0.011	0.005	0.002
19.00	6.69	67.4	70	0.730	0.170	0.009	0.072	0.012	0.006	0.002
21.00	6.83	64.8	66	0.740	0.169	0.009	0.064	0.012	0.005	0.001

Table 57

Composition of Rumen Contents for Sheep No. 437 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.41	92.2	98	0.726	0.150	0.008	0.096	0.012	0.007	0.001
10.00	5.53	140.5	136	0.651	0.203	0.006	0.126	0.008	0.006	0.001
11.00	5.98	159.7	157	0.626	0.216	0.006	0.134	0.008	0.009	0.001
12.00	5.66	167.8	179	0.623	0.263	0.006	0.093	0.006	0.009	0.001
13.00	5.60	164.7	152	0.665	0.187	0.005	0.126	0.007	0.010	0.002
15.00	6.04	111.8	106	0.683	0.176	0.006	0.116	0.008	0.009	0.001
17.00	6.15	120.0	89	0.689	0.168	0.006	0.113	0.014	0.009	0.002
19.00	6.35	112.0	76	0.718	0.156	0.009	0.098	0.010	0.007	0.002
21.00	6.34	109.0	77	0.726	0.154	0.008	0.093	0.011	0.007	0.002

Table 58

Composition of Rumen Contents for Sheep No. 447 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.14	25.1	82	0.708	0.169	0.028	0.059	0.029	0.006	0.002
10.00	7.00	37.8	103	0.629	0.196	0.024	0.098	0.035	0.012	0.006
11.00	7.02	30.7	90	0.655	0.181	0.019	0.097	0.037	0.012	0.003
12.00	7.00	36.8	86	0.653	0.185	0.021	0.091	0.035	0.011	0.004
13.00	7.01	35.9	90	0.669	0.175	0.019	0.090	0.033	0.010	0.005
15.00	7.12	32.8	73	0.693	0.168	0.024	0.073	0.032	0.008	0.003
17.00	7.15	26.2	69	0.703	0.167	0.023	0.064	0.034	0.008	0.002
19.00	7.24	23.6	86	0.696	0.162	0.028	0.068	0.035	0.008	0.002
21.00	7.33	20.7	82	0.707	0.162	0.031	0.053	0.040	0.007	0.002

Table 59

Composition of Rumen Contents for Sheep No. 447 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.87	49.8	122	0.702	0.192	0.013	0.072	0.014	0.006	0.001
10.00	6.10	104.7	223	0.555	0.290	0.009	0.124	0.010	0.009	0.003
11.00	6.25	104.7	220	0.590	0.264	0.011	0.110	0.009	0.012	0.004
12.00	6.46	103.9	170	0.631	0.238	0.009	0.101	0.007	0.011	0.004
13.00	6.35	88.8	136	0.651	0.228	0.008	0.095	0.006	0.009	0.003
15.00	6.67	70.0	80	0.672	0.215	0.010	0.086	0.007	0.008	0.002
17.00	6.86	65.2	80	0.701	0.198	0.010	0.073	0.008	0.007	0.003
19.00	6.83	57.3	114	0.710	0.195	0.012	0.064	0.011	0.006	0.002
21.00	7.00	44.2	109	0.686	0.209	0.013	0.071	0.014	0.006	0.001

Table 60

Composition of Rumen Contents for Sheep No. 447 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.83	73.4	87	0.659	0.180	0.012	0.127	0.013	0.007	0.002
10.00	5.70	130.2	181	0.584	0.241	0.005	0.156	0.008	0.005	0.002
11.00	6.00	148.8	222	0.563	0.276	0.005	0.141	0.006	0.008	0.002
12.00	6.14	138.7	188	0.574	0.263	0.005	0.141	0.006	0.010	0.002
13.00	6.12	132.2	114	0.608	0.234	0.005	0.134	0.005	0.010	0.002
15.00	6.51	106.5	66	0.636	0.207	0.010	0.132	0.005	0.008	0.002
17.00	6.65	97.5	74	0.656	0.195	0.006	0.129	0.005	0.007	0.003
19.00	6.84	84.7	74	0.659	0.189	0.007	0.128	0.008	0.007	0.002
21.00	6.88	68.8	87	0.668	0.180	0.012	0.121	0.010	0.007	0.002

Table 61

Composition of Rumen Contents for Sheep No. 448 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.04	54.4	75	0.720	0.163	0.011	0.084	0.014	0.006	0.002
10.00	6.43	86.3	183	0.589	0.231	0.009	0.142	0.016	0.011	0.002
11.00	6.44	90.7	162	0.597	0.217	0.011	0.137	0.018	0.012	0.008
12.00	6.47	87.7	157	0.640	0.197	0.009	0.121	0.016	0.011	0.008
13.00	6.61	87.2	122	0.667	0.178	0.010	0.120	0.012	0.009	0.005
15.00	6.74	73.2	74	0.703	0.175	0.009	0.090	0.012	0.008	0.003
17.00	6.95	64.7	65	0.726	0.164	0.010	0.082	0.011	0.006	0.002
19.00	7.07	53.8	87	0.726	0.165	0.013	0.073	0.015	0.006	0.002
21.00	7.15	47.1	57	0.728	0.174	0.012	0.064	0.015	0.005	0.002

Table 62

Composition of Rumen Contents for Sheep No. 448 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.92	60.6	97	0.712	0.190	0.012	0.068	0.011	0.006	0.002
10.00	5.95	120.0	199	0.577	0.300	0.006	0.098	0.010	0.007	0.002
11.00	6.01	133.6	241	0.574	0.294	0.008	0.100	0.010	0.011	0.003
12.00	6.43	115.0	229	0.603	0.266	0.010	0.095	0.010	0.013	0.004
13.00	6.44	98.1	170	0.624	0.249	0.008	0.096	0.009	0.011	0.003
15.00	6.60	87.8	89	0.667	0.219	0.008	0.088	0.007	0.008	0.003
17.00	6.72	61.2	76	0.702	0.195	0.011	0.071	0.013	0.007	0.002
19.00	7.01	54.4	76	0.702	0.198	0.009	0.078	0.008	0.005	0.002
21.00	6.98	52.5	68	0.711	0.195	0.009	0.071	0.008	0.005	0.001

Table 63

Composition of Rumen Contents for Sheep No. 448 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.71	61.1	94	0.678	0.170	0.012	0.124	0.011	0.006	0.001
10.00	5.92	97.6	146	0.586	0.263	0.006	0.130	0.007	0.008	0.001
11.00	6.19	102.3	164	0.594	0.250	0.005	0.133	0.006	0.011	0.002
12.00	6.29	99.4	126	0.616	0.227	0.005	0.135	0.005	0.011	0.001
13.00	6.18	104.8	95	0.636	0.211	0.004	0.134	0.004	0.011	0.001
15.00	6.44	104.2	48	0.666	0.190	0.004	0.126	0.004	0.008	0.002
17.00	6.71	80.6	61	0.669	0.185	0.005	0.126	0.006	0.007	0.002
19.00	6.93	73.9	99	0.683	0.170	0.006	0.125	0.009	0.007	0.001
21.00	7.13	66.5	112	0.682	0.173	0.011	0.115	0.012	0.006	0.001

Table 64

Composition of Rumen Contents for Sheep No. 449 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.25	36.1	101	0.716	0.175	0.024	0.048	0.029	0.007	0.001
10.00	6.68	71.5	186	0.541	0.270	0.013	0.137	0.025	0.010	0.003
11.00	6.69	83.9	186	0.556	0.249	0.015	0.129	0.034	0.013	0.004
12.00	6.71	81.4	179	0.587	0.227	0.018	0.116	0.031	0.013	0.008
13.00	6.73	70.8	102	0.628	0.210	0.014	0.103	0.032	0.010	0.005
15.00	6.87	59.4	72	0.659	0.197	0.015	0.079	0.040	0.008	0.002
17.00	7.01	59.5	72	0.712	0.186	0.012	0.068	0.016	0.005	0.002
19.00	7.01	51.2	81	0.720	0.176	0.019	0.056	0.022	0.006	0.001
21.00	7.16	33.6	82	0.712	0.179	0.021	0.056	0.026	0.007	0.001

Table 65

Composition of Rumen Contents for Sheep No. 449 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.09	42.7	95	0.729	0.177	0.014	0.060	0.013	0.006	0.001
10.00	6.52	80.7	198	0.593	0.266	0.010	0.104	0.015	0.008	0.004
11.00	6.50	85.9	206	0.616	0.245	0.011	0.099	0.012	0.011	0.005
12.00	6.58	81.3	159	0.637	0.226	0.010	0.095	0.015	0.011	0.006
13.00	6.64	66.3	103	0.660	0.212	0.010	0.091	0.009	0.011	0.006
15.00	6.80	65.4	52	0.705	0.193	0.009	0.076	0.007	0.008	0.002
17.00	6.98	60.0	61	0.727	0.181	0.009	0.067	0.007	0.007	0.003
19.00	7.11	49.7	86	0.723	0.181	0.011	0.066	0.011	0.007	0;002
21.00	7.14	39.4	103	0.716	0.180	0.015	0.067	0.014	0.006	0.002

Table 66

Composition of Rumen Contents for Sheep No. 449 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.82	75.0	148	0.694	0.171	0.012	0.103	0.011	0.007	0.002
10.00	5.83	104.8	196	0.640	0.202	0.006	0.138	0.007	0.006	0.002
11.00	6.26	141.0	244	0.575	0.252	0.006	0.148	0.007	0.011	0.001
12.00	6.35	114.7	210	0.590	0.238	0.006	0.145	0.007	0.013	0.001
13.00	6.36	106.6	153	0.603	0.225	0.006	0.144	0.007	0.013	0.002
15.00	6.43	110.4	87	0.635	0.198	0.007	0.140	0.007	0.011	0.002
17.00	6.79	98.3	101	0.680	0.189	0.008	0.106	0.008	0.009	0.001
19.00	6.76	82.2	61	0.694	0.182	0.006	0.099	0.011	0.008	0.001
21.00	6.87	67.9	96	0.692	0.182	0.011	0.094	0.012	0.007	0.001

Table 67

Composition of Rumen Contents for Sheep No. 680 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.89	70.9	74	0.776	0.146	0.009	0.053	0.010	0.005	0.001
10.00	6.36	86.7	205	0.682	0.193	0.007	0.103	0.009	0.004	0.002
11.00	6.21	102.9	231	0.675	0.172	0.009	0.101	0.026	0.011	0.007
12.00	6.26	96.3	148	0.709	0.158	0.008	0.087	0.022	0.009	0.005
13.00	6.32	91.8	92	0.753	0.146	0.007	0.068	0.016	0.007	0.003
15.00	6.60	77.3	70	0.796	0.129	0.006	0.051	0.011	0.004	0.001
17.00	6.64	77.5	70	0.808	0.127	0.006	0.045	0.009	0.003	0.001
19.00	6.67	74.2	74	0.810	0.127	0.007	0.043	0.010	0.004	0.001
21.00	6.81	60.5	74	0.812	0.128	0.009	0.037	0.011	0.004	0.001

Table 68

Composition of Rumen Contents for Sheep No. 680 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.87	55.9	99	0.733	0.162	0.012	0.074	0.013	0.006	0.002
10.00	6.34	84.6	168	0.620	0.213	0.010	0.122	0.014	0.014	0.006
11.00	6.27	88.1	198	0.629	0.203	0.009	0.123	0.014	0.014	0.009
12.00	6.36	85.6	151	0.670	0.188	0.010	0.109	0.011	0.005	0.007
13.00	6.47	73.0	95	0.696	0.176	0.007	0.099	0.009	0.010	0.004
15.00	6.73	66.3	65	0.727	0.167	0.008	0.083	0.007	0.007	0.002
17.00	6.83	59.8	77	0.740	0.160	0.008	0.077	0.007	0.006	0.002
19.00	7.00	55.9	73	0.732	0.161	0.011	0.077	0.010	0.006	0.002
21.00	6.90	54.3	82	0.731	0.164	0.011	0.074	0.011	0.006	0.002

Table 69

Composition of Rumen Contents for Sheep No. 680 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.58	94.2	93	0.658	0.234	0.010	0.078	0.012	0.007	0.001
10.00	5.30	136.2	165	0.603	0.308	0.005	0.071	0.006	0,006	0.000
11.00	5.50	180.4	254	0.531	0.372	0.004	0.080	0.006	0.007	0.000
12.00	5.88	171.4	249	0.589	0.288	0.004	0.103	0.006	0.009	0.001
13.00	5.87	161.5	233	0.560	0.333	0.005	0.085	0.007	0.010	0.001
15.00	6.18	128.0	166	0.579	0.308	0.005	0.087	0.010	0.010	0.001
17.00	6.35	117.2	119	0.626	0.267	0.006	0.084	0.007	0.009	0.001
19.00	6.46	113.2	93	0.650	0.245	0.007	0.080	0.010	0.008	0.001
21.00	6.53	91.3	72	0.655	0.239	0.008	0.077	0.011	0.007	0.002

Table 70

Composition of Blood from Individual Sheep

Metabolite	Treatment	Sheep No.								
		409	414	434	435	437	447	448	449	680
Blood pH	A	7.34	7.33	7.30	7.17	7.36	7.21	7.35	7.38	7.33
	B	7.40	7.27	7.30	7.33	7.36	7.32	7.42	7.35	7.40
	C	7.32	7.33	7.34	7.31	7.38	7.41	7.41	7.35	7.37
Plasma glucose mg/l	A	494	629	534	632	528	500	509	553	528
	B	466	581	538	659	513	623	629	494	459
	C	581	547	628	616	596	509	488	503	532
Plasma urea-nitrogen mg/l	A	117	105	112	136	127	146	127	134	146
	B	103	122	93	122	84	114	78	160	88
	C	112	103	98	108	83	117	98	88	85

A = Control silage
B = Treated silage
C = Dried grass

Table 71

Nutritive Value of Silages and Dried Grass for Individual Sheep

	Treatment	Sheep No.								
		409	414	434	435	437	443	447	448	449
Daily intake $g/kgW^{0.75}$	A	16.8	26.2	31.8	17.2	19.4	22.8	12.9	13.0	21.2
	B	17.3	16.5	37.0	38.7	22.6	26.8	19.7	40.3	26.7
	C	34.2	37.0	33.3	29.9	38.4	23.1	33.4	35.8	30.3
Daily intake g/day	A	395	553	746	294	426	551	265	272	459
	B	398	332	877	704	482	584	465	873	586
	C	820	788	766	529	815	478	726	770	669
Dry matter digestibility	A	0.774	0.784	0.764	0.790	0.787	0.728	0.817	0.753	0.677
	B	0.733	0.764	0.694	0.706	0.713	-	0.713	0.666	0.743
	C	0.705	0.725	0.704	0.713	0.704	0.726	0.706	0.700	0.680
Organic matter digestibility	A	0.810	0.821	0.795	0.830	0.812	0.745	0.862	0.794	0.786
	B	0.778	0.800	0.731	0.746	0.759	-	0.760	0.711	0.734
	C	0.753	0.758	0.728	0.747	0.744	0.754	0.724	0.740	0.737
Nitrogen digestibility	A	0.852	0.855	0.841	0.854	0.862	0.809	0.878	0.805	0.821
	B	0.810	0.790	0.803	0.779	0.798	-	0.808	0.767	0.782
	C	0.803	0.802	0.817	0.792	0.801	0.812	0.804	0.805	0.788
Metabolisable energy MJ/kg	A	13.6	15.0	14.0	13.6	14.4	13.2	14.3	13.1	13.3
	B	11.7	11.8	11.6	10.2	11.7	-	11.3	10.5	10.5
	C	10.3	10.4	10.3	10.8	10.2	10.1	10.4	10.0	9.9

Table 71 (Cont'd)

Nutritive Value of Silages and Dried Grass for Individual Sheep

	Treat-ment	Sheep No.								
		409	414	434	435	437	443	447	448	449
Gross energy of digestible organic matter MJ/kg	A	24.3	24.3	24.4	24.3	24.4	24.5	24.0	24.4	24.4
	B	22.1	22.2	22.3	22.0	21.9	-	22.3	22.0	22.2
	C	19.6	19.7	20.1	19.5	19.7	19.8	20.1	19.4	19.5

A = Control silage
B = Treated silage
C = Dried grass

Table 72

Composition of Rumen Contents of Sheep No. 409 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.05	51.4	75	0.732	0.170	0.022	0.047	0.023	0.006	0.001
10.00	6.87	97.0	281	0.651	0.230	0.020	0.052	0.030	0.014	0.003
11.00	6.82	84.9	328	0.647	0.211	0.020	0.076	0.029	0.014	0.004
12.00	6.80	81.2	258	0.684	0.197	0.018	0.063	0.025	0.011	0.003
13.00	6.86	72.6	178	0.697	0.189	0.021	0.057	0.024	0.009	0.002
15.00	7.03	59.2	122	0.715	0.180	0.022	0.054	0.022	0.007	0.001
17.00	7.15	53.5	159	0.730	0.170	0.023	0.048	0.022	0.006	0.001
19.00	7.20	52.2	173	0.733	0.169	0.024	0.046	0.024	0.005	0.001
21.00	7.28	47.5	150	0.737	0.158	0.026	0.044	0.028	0.006	0.001

Table 73

Composition of Rumen Contents of Sheep No. 409 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.16	50.9	114	0.739	0.156	0.025	0.049	0.025	0.005	0.001
10.00	6.95	52.1	294	0.684	0.159	0.022	0.076	0.038	0.014	0.007
11.00	6.93	73.5	368	0.706	0.140	0.024	0.070	0.037	0.014	0.010
12.00	6.90	77.1	274	0.704	0.147	0.025	0.072	0.033	0.014	0.006
13.00	6.95	76.0	233	0.725	0.143	0.023	0.067	0.029	0.010	0.003
15.00	6.99	60.6	123	0.729	0.151	0.021	0.064	0.026	0.008	0.002
17.00	7.06	57.1	102	0.732	0.155	0.021	0.060	0.024	0.006	0.002
19.00	7.14	52.4	86	0.746	0.150	0.021	0.053	0.024	0.005	0.001
21.00	7.25	48.8	82	0.753	0.147	0.021	0.048	0.025	0.005	0.001

Table 74

Composition of Rumen Contents of Sheep No. 409 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.84	91.4	256	0.726	0.172	0.016	0.059	0.020	0.006	0.001
10.00	6.64	118.0	449	0.741	0.158	0.015	0.060	0.017	0.008	0.001
11.00	6.62	116.6	483	0.729	0.159	0.015	0.067	0.021	0.008	0.001
12.00	6.53	124.7	470	0.717	0.171	0.014	0.070	0.019	0.009	0.001
13.00	6.39	132.4	372	0.721	0.174	0.012	0.070	0.014	0.008	0.002
15.00	6.38	130.0	265	0.734	0.169	0.012	0.065	0.012	0.007	0.001
17.00	6.27	125.9	260	0.734	0.170	0.012	0.062	0.013	0.007	0.001
19.00	6.47	107.3	214	0.736	0.169	0.013	0.060	0.014	0.006	0.001
21.00	6.78	95.6	248	0.723	0.173	0.016	0.060	0.019	0.006	0.001

Table 75

Composition of Rumen Contents of Sheep No. 414 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.16	48.3	122	0.702	0.197	0.021	0.051	0.023	0.005	0.001
10.00	6.93	88.9	267	0.644	0.222	0.020	0.059	0.037	0.014	0.004
11.00	6.84	90.9	337	0.641	0.221	0.021	0.063	0.033	0.016	0.004
12.00	6.80	91.2	304	0.664	0.211	0.019	0.063	0.028	0.013	0.003
13.00	6.93	54.9	229	0.667	0.212	0.018	0.064	0.027	0.010	0.002
15.00	7.01	51.3	150	0.696	0.197	0.019	0.060	0.021	0.006	0.002
17.00	7.05	50.3	117	0.709	0.193	0.016	0.056	0.019	0.006	0.001
19.00	7.19	49.2	136	0.715	0.187	0.021	0.051	0.021	0.005	0.001
21.00	7.24	45.7	104	0.718	0.182	0.023	0.047	0.025	0.006	0.000

Table 76

Composition of Rumen Contents of Sheep No. 414 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.16	41.0	131	0.727	0.175	0.024	0.047	0.020	0.005	0.002
10.00	7.06	45.3	254	0.653	0.162	0.029	0.091	0.041	0.016	0.010
11.00	7.01	59.0	293	0.665	0.149	0.028	0.090	0.037	0.017	0.016
12.00	7.08	59.7	237	0.694	0.146	0.027	0.079	0.030	0.015	0.008
13.00	7.04	62.9	192	0.711	0.154	0.024	0.071	0.027	0.010	0.004
15.00	7.09	55.3	147	0.714	0.165	0.024	0.063	0.024	0.008	0.003
17.00	7.15	48.6	110	0.718	0.165	0.024	0.058	0.024	0.007	0.003
19.00	7.24	46.2	131	0.727	0.165	0.024	0.052	0.024	0.006	0.002
21.00	7.28	35.6	110	0.735	0.161	0.024	0.047	0.026	0.006	0.002

Table 77

Composition of Rumen Contents of Sheep No. 414 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.84	69.8	222	0.727	0.157	0.021	0.063	0.025	0.007	0.001
10.00	6.65	105.4	457	0.732	0.159	0.017	0.062	0.021	0.008	0.001
11.00	6.60	93.5	445	0.704	0.169	0.016	0.078	0.021	0.011	0.002
12.00	6.57	104.7	359	0.710	0.168	0.016	0.074	0.020	0.010	0.002
13.00	6.65	90.1	270	0.725	0.159	0.015	0.078	0.015	0.009	0.000
15.00	6.62	88.6	201	0.729	0.160	0.013	0.073	0.016	0.007	0.001
17.00	6.62	83.8	146	0.733	0.160	0.014	0.067	0.017	0.007	0.002
19.00	6.82	81.3	188	0.731	0.159	0.018	0.065	0.021	0.006	0.001
21.00	6.98	68.2	226	0.730	0.155	0.020	0.062	0.025	0.007	0.001

Table 78

Composition of Rumen Contents of Sheep No. 434 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.09	55.6	154	0.720	0.182	0.017	0.054	0.020	0.005	0.001
10.00	6.67	95.5	325	0.685	0.197	0.017	0.058	0.025	0.016	0.004
11.00	6.62	103.4	449	0.664	0.219	0.017	0.054	0.025	0.017	0.004
12.00	6.53	105.8	394	0.681	0.199	0.018	0.063	0.021	0.015	0.004
13.00	6.49	99.5	304	0.691	0.197	0.015	0.065	0.018	0.011	0.003
15.00	6.67	72.9	159	0.709	0.191	0.014	0.065	0.013	0.007	0.002
17.00	6.71	58.5	120	0.717	0.191	0.013	0.059	0.013	0.006	0.002
19.00	6.82	53.9	98	0.727	0.188	0.013	0.053	0.013	0.005	0.001
21.00	6.98	47.0	129	0.720	0.191	0.016	0.051	0.016	0.006	0.001

Table 79

Composition of Rumen Contents of Sheep No. 434 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.88	66.2	90	0.730	0.169	0.013	0.065	0.016	0.006	0.001
10.00	6.51	90.1	413	0.699	0.163	0.015	0.074	0.025	0.015	0.009
11.00	6.47	120.4	527	0.703	0.151	0.017	0.075	0.026	0.017	0.014
12.00	6.21	122.0	511	0.712	0.145	0.018	0.068	0.024	0.020	0.010
13.00	6.55	107.5	368	0.728	0.149	0.015	0.067	0.021	0.014	0.006
15.00	6.53	98.5	262	0.739	0.157	0.013	0.066	0.016	0.008	0.004
17.00	6.51	85.2	184	0.735	0.161	0.012	0.066	0.016	0.008	0.002
19.00	6.59	75.9	127	0.727	0.172	0.012	0.065	0.014	0.007	0.002
21.00	6.73	73.9	127	0.749	0.161	0.011	0.059	0.014	0.006	0.001

Table 80

Composition of Rumen Contents of Sheep No. 434 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.86	71.1	187	0.684	0.196	0.017	0.071	0.022	0.008	0.002
10.00	6.69	118.9	454	0.695	0.197	0.014	0.064	0.020	0.008	0.001
11.00	6.55	122.1	533	0.690	0.204	0.014	0.063	0.018	0.009	0.001
12.00	6.45	107.4	501	0.671	0.217	0.013	0.070	0.016	0.011	0.002
13.00	6.33	134.7	407	0.668	0.219	0.013	0,074	0.014	0.011	0.002
15.00	6.35	117.6	295	0.664	0.225	0.012	0.073	0.014	0.009	0.002
17.00	6.35	108.3	239	0.679	0.216	0.014	0.070	0.013	0.009	0.002
19.00	6.64	95.6	253	0.670	0.212	0.015	0.075	0.018	0.008	0.002
21.00	6.86	86.3	215	0.679	0.205	0.017	0.071	0.020	0.008	0,002

Table 81

Composition of Rumen Contents of Sheep No. 435 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.95	41.6	163	0.722	0.169	0.025	0.047	0.029	0.007	0.001
10.00	6.73	60.8	343	0.630	0.226	0.025	0.053	0.044	0.018	0.004
11.00	6.61	77.4	331	0.642	0.213	0.021	0.066	0.034	0.016	0.007
12.00	6.61	63.5	262	0.668	0.202	0.023	0.059	0.031	0.014	0.004
13.00	6.67	60.2	237	0.674	0.201	0.022	0.059	0.029	0.013	0.002
15.00	6.67	50.4	184	0.704	0.185	0.019	0.057	0.024	0.010	0.002
17.00	6.82	47.0	151	0.707	0.179	0.022	0.053	0.028	0.008	0.002
19.00	7.00	41.4	110	0.717	0.177	0.022	0.047	0.028	0.008	0.002
21.00	7.02	42.2	139	0.730	0.168	0.022	0.045	0.026	0.008	0.001

Table 82

Composition of Rumen Contents of Sheep No. 435 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.07	60.3	111	0.744	0.156	0.016	0.063	0.014	0.006	0.001
10.00	6.80	91.7	436	0.694	0.134	0.020	0.099	0.030	0.012	0.010
11.00	6.77	93.6	445	0.700	0.131	0.020	0.093	0.029	0.017	0.010
12.00	6.56	108.9	355	0.703	0.134	0.020	0.091	0.026	0.017	0.010
13.00	6.63	86.8	252	0.703	0.142	0.020	0.090	0.025	0.014	0.007
15.00	6.74	78.5	133	0.729	0.150	0.016	0.077	0.016	0.009	0.004
17.00	6.81	62.7	133	0.736	0.155	0.015	0.070	0.015	0.007	0.003
19.00	6.92	61.8	86	0.739	0.158	0.014	0.065	0.015	0.006	0.003
21.00	7.02	55.6	65	0.742	0.160	0.014	0.060	0.015	0.006	0.003

Table 83

Composition of Rumen Contents of Sheep No. 435 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.75	104.7	243	0.744	0.158	0.014	0.056	0.021	0.006	0.001
10.00	6.40	127.0	440	0.725	0.167	0.013	0.067	0.019	0.008	0.002
11.00	6.32	128.7	468	0.723	0.168	0.013	0.069	0.017	0.009	0.002
12.00	6.13	165.4	492	0.724	0.171	0.011	0.069	0.015	0.009	0.002
13.00	6.25	139.2	402	0.721	0.171	0.013	0.069	0.015	0.009	0.002
15.00	6.07	133.5	342	0.732	0.168	0.009	0.067	0.015	0.009	0.002
17.00	6.48	111.2	239	0.748	0.160	0.012	0.057	0.015	0.007	0.001
19.00	6.63	90.7	248	0.747	0.156	0.015	0.056	0.018	0.007	0.001
21.00	6.78	86.3	201	0.746	0.155	0.017	0.053	0.021	0.006	0.001

Table 84

Composition of Rumen Contents of Sheep No. 437 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.01	41.7	145	0.673	0.203	0.025	0.064	0.026	0.007	0.001
10.00	6.83	56.7	248	0.664	0.213	0.021	0.058	0.029	0.013	0.003
11.00	6.78	63.7	270	0.664	0.207	0.021	0.063	0.029	0.012	0.004
12.00	6.77	59.6	235	0.664	0.206	0.021	0.068	0.027	0.012	0.003
13.00	6.90	50.5	205	0.662	0.205	0.024	0.068	0.028	0.011	0.002
15.00	6.91	46.6	154	0.679	0.202	0.021	0.064	0.024	0.009	0.001
17.00	6.90	40.3	150	0.679	0.204	0.022	0.061	0.025	0.008	0.001
19.00	6.91	37.8	137	0.677	0.205	0.022	0.062	0.025	0.007	0.001
21.00	6.93	36.9	119	0.677	0.206	0.023	0.061	0.025	0.007	0.000

Table 85

Composition of Rumen Contents of Sheep No. 437 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.69	84.5	187	0.750	0.172	0.014	0.042	0.018	0.004	0.000
10.00	6.67	98.8	346	0.721	0.157	0.017	0.063	0.027	0.010	0.005
11.00	6.59	100.0	398	0.727	0.145	0.018	0.065	0.028	0.010	0.006
12.00	6.35	107.0	379	0.738	0.146	0.016	0.062	0.024	0.009	0.005
13.00	6.55	89.9	328	0.737	0.152	0.018	0.061	0.022	0.007	0.003
15.00	6.58	80.4	234	0.748	0.157	0.015	0.055	0.018	0.006	0.002
17.00	6.65	74.8	182	0.756	0.161	0.014	0.048	0.016	0.004	0.001
19.00	6.83	72.6	187	0.762	0.163	0.014	0.042	0.015	0.003	0.000
21.00	6.80	72.4	159	0.772	0.161	0.012	0.040	0.015	0.003	0.000

Table 86

Composition of Rumen Contents of Sheep No. 437 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.68	79.9	310	0.730	0.167	0.017	0.053	0.026	0.006	0.001
10.00	6.53	87.6	515	0.699	0.179	0.016	0.071	0.025	0.009	0.001
11.00	6.33	94.6	494	0.709	0.173	0.015	0.071	0.020	0.009	0.002
12.00	6.31	102.4	401	0.722	0.171	0.014	0.068	0.016	0.009	0.001
13.00	6.35	103.3	347	0.722	0.172	0.014	0.066	0.015	0.008	0.001
15.00	6.13	96.6	327	0.726	0.172	0.012	0.067	0.015	0.008	0.001
17.00	6.22	88.3	270	0.725	0.172	0.013	0.063	0.019	0.008	0.001
19.00	6.50	81.7	258	0.724	0.171	0.015	0.060	0.021	0.008	0.001
21.00	6.74	72.0	253	0.730	0.167	0.017	0.055	0.023	0.007	0.001

Table 87

Composition of Rumen Contents of Sheep No. 443 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.22	42.6	188	0.723	0.178	0.021	0.041	0.029	0.008	0.001
10.00	6.70	79.7	402	0.639	0.232	0.021	0.050	0.034	0.020	0.004
11.00	6.79	90.8	402	0.642	0.223	0.020	0.062	0.031	0.017	0.005
12.00	6.79	67.5	295	0.657	0.211	0.022	0.061	0.029	0.015	0.004
13.00	6.78	62.2	241	0.671	0.202	0.022	0.061	0.028	0.014	0.003
15.00	6.85	51.8	141	0.704	0.192	0.020	0.050	0.022	0.010	0.002
17.00	6.95	48.6	137	0.738	0.181	0.019	0.034	0.020	0.006	0.001
19.00	6.92	46.2	132	0.743	0.173	0.018	0.038	0.021	0.007	0.001
21.00	6.98	42.7	127	0.739	0.179	0.019	0.035	0.022	0.007	0.001

Table 88

Composition of Rumen Contents of Sheep No. 680* on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.01	51.8	172	0.723	0.170	0.022	0.050	0.028	0.007	0.002
10.00	6.80	75.0	429	0.676	0.156	0.024	0.092	0.032	0.015	0.005
11.00	6.59	87.9	442	0.689	0.139	0.022	0.093	0.028	0.017	0.012
12.00	6.66	78.1	458	0.704	0.140	0.022	0.089	0.025	0.013	0.008
13.00	6.60	69.6	409	0.730	0.141	0.019	0.075	0.020	0.009	0.005
15.00	6.73	59.9	217	0.727	0.152	0.018	0.074	0.018	0.007	0.003
17.00	6.82	55.8	176	0.733	0.156	0.018	0.067	0.018	0.007	0.002
19.00	6.90	50.4	164	0.736	0.153	0.019	0.064	0.021	0.007	0.002
21.00	7.01	46.4	192	0.733	0.159	0.020	0.058	0.022	0.007	0.001

*replacing sheep No. 443.

Table 89

Composition of Rumen Contents of Sheep No. 68* on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.71	75.8	229	0.770	0.140	0.017	0.050	0.017	0.006	0.001
10.00	6.64	88.5	470	0.758	0.148	0.018	0.040	0.026	0.010	0.001
11.00	6.62	100.4	491	0.753	0.156	0.014	0.047	0.019	0.009	0.002
12.00	6.60	133.5	540	0.750	0.156	0.014	0.054	0.016	0.011	0.001
13.00	6.52	119.7	462	0.739	0.156	0.014	0.064	0.016	0.010	0.001
15.00	6.56	101.1	270	0.746	0.159	0.013	0.055	0.015	0.009	0.001
17.00	6.73	91.2	192	0.748	0.163	0.015	0.051	0.017	0.007	0.001
19.00	6.63	79.9	172	0.754	0.160	0.015	0.046	0.019	0.007	0.001
21.00	6.82	72.9	164	0.755	0.149	0.016	0.048	0.023	0.007	0.001

*replacing Sheep No. 443.

Table 90

Composition of Rumen Contents of Sheep No. 447 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.25	34.6	103	0.712	0.183	0.028	0.044	0.028	0.006	0.001
10.00	7.05	55.0	267	0.636	0.235	0.026	0.045	0.038	0.016	0.004
11.00	7.02	55.6	244	0.635	0.230	0.026	0.052	0.037	0.016	0.004
12.00	7.01	51.5	187	0.653	0.219	0.027	0.055	0.032	0.012	0.003
13.00	6.98	43.8	131	0.655	0.216	0.027	0.058	0.032	0.010	0.003
15.00	7.10	35.5	99	0.684	0.200	0.027	0.051	0.028	0.009	0.001
17.00	7.13	31.8	70	0.699	0.197	0.030	0.040	0.027	0.007	0.001
19.00	7.18	31.9	98	0.701	0.191	0.031	0.040	0.030	0.007	0.001
21.00	7.34	27.4	103	0.705	0.184	0.032	0.041	0.032	0.006	0.000

Table 91

Composition of Rumen Contents of Sheep No. 447 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.18	59.6	154	0.728	0.166	0.024	0.052	0.024	0.005	0.001
10.00	6.80	83.0	406	0.681	0.129	0.026	0.108	0.030	0.014	0.012
11.00	6.75	90.9	351	0.686	0.123	0.025	0.113	0.027	0.015	0.011
12.00	6.71	91.2	304	0.696	0.129	0.023	0.106	0.025	0.013	0.009
13.00	6.64	80.0	218	0.707	0.140	0.021	0.095	0.021	0.011	0.005
15.00	6.80	64.9	120	0.729	0.158	0.020	0.074	0.010	0.007	0.003
17.00	6.87	55.2	94	0.729	0.157	0.020	0.070	0.016	0.005	0.001
19.00	6.95	46.1	107	0.730	0.170	0.019	0.056	0.019	0.005	0.001
21.00	7.05	45.2	151	0.747	0.167	0.018	0.044	0.020	0.005	0.001

Table 92

Composition of Rumen Contents of Sheep No. 447 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.83	81.2	180	0.727	0.186	0.018	0.039	0.022	0.008	0.001
10.00	6.60	101.7	560	0.709	0.193	0.016	0.044	0.026	0.010	0.002
11.00	6.59	114.2	585	0.715	0.182	0.015	0.054	0.020	0.012	0.003
12.00	6.59	116.8	454	0.726	0.173	0.014	0.055	0.015	0.012	0.002
13.00	6.58	112.9	384	0.722	0.182	0.014	0.056	0.015	0.011	0.002
15.00	6.49	107.3	245	0.722	0.190	0.012	0.053	0.013	0.009	0.001
17.00	6.70	94.9	184	0.717	0.197	0.015	0.048	0.015	0.008	0.001
19.00	6.66	88.7	200	0.734	0.189	0.015	0.034	0.019	0.009	0.002
21.00	6.95	75.4	196	0.743	0.182	0.017	0.029	0.021	0.008	0.001

Table 93

Composition of Rumen Contents of Sheep No. 448 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.09	35.8	188	0.686	0.195	0.027	0.056	0.029	0.006	0.001
10.00	6.91	65.6	458	0.641	0.224	0.024	0.053	0.040	0.015	0.004
11.00	6.82	74.2	401	0.633	0.222	0.024	0.057	0.038	0.018	0.008
12.00	6.85	68.5	348	0.637	0.219	0.025	0.063	0.033	0.018	0.005
13.00	6.79	63.4	311	0.659	0.214	0.023	0.060	0.027	0.014	0.004
15.00	6.74	61.2	217	0.665	0.214	0.020	0.059	0.027	0.012	0.003
17.00	6.93	53.5	147	0.692	0.205	0.019	0.051	0.023	0.007	0.002
19.00	6.98	49.6	143	0.693	0.206	0.019	0.050	0.022	0.007	0.002
21.00	7.07	45.2	139	0.696	0.205	0.021	0.047	0.024	0.007	0.001

Table 94

Composition of Rumen Contents of Sheep No. 448 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.15	67.8	120	0.738	0.162	0.018	0.057	0.018	0.005	0.002
10.00	6.67	96.4	449	0.688	0.140	0.018	0.102	0.027	0.013	0.012
11.00	6.56	106.3	470	0.693	0.136	0.021	0.093	0.028	0.017	0.012
12.00	6.50	116.6	428	0.695	0.144	0.019	0.091	0.023	0.018	0.010
13.00	6.43	113.1	343	0.712	0.141	0.018	0.085	0.021	0.015	0.008
15.00	6.54	87.3	171	0.724	0.153	0.016	0.075	0.017	0.009	0.005
17.00	6.69	74.4	120	0.734	0.156	0.014	0.067	0.014	0.008	0.006
19.00	6.87	58.2	111	0.736	0.164	0.015	0.061	0.015	0.007	0.003
21.00	7.10	56.5	118	0.741	0.165	0.015	0.055	0.016	0.006	0.002

Table 95

Composition of Rumen Contents of Sheep No. 448 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.76	90.0	253	0.717	0.168	0.018	0.067	0.023	0.006	0.001
10.00	6.68	117.6	487	0.733	0.156	0.015	0.069	0.019	0.007	0.002
11.00	6.58	118.8	515	0.735	0.159	0.015	0.064	0.017	0.009	0.002
12.00	6.58	121.7	477	0.725	0.163	0.015	0.070	0.017	0.009	0.002
13.00	6.40	133.2	449	0.718	0.166	0.014	0.075	0.016	0.010	0.002
15.00	6.40	116.9	304	0.718	0.170	0.015	0.072	0.016	0.009	0.002
17.00	6.50	112.7	295	0.725	0.168	0.015	0.070	0.017	0.007	0.001
19.00	6.69	96.2	229	0.726	0.166	0.016	0.065	0.019	0.007	0.002
21.00	6.85	93.3	215	0.729	0.166	0.018	0.060	0.020	0.007	0.001

Table 96

Composition of Rumen Contents of Sheep No. 449 on Control Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.01	42.7	147	0.720	0.185	0.024	0.035	0.028	0.006	0.002
10.00	6.99	64.9	339	0.634	0.205	0.026	0.064	0.043	0.018	0.010
11.00	6.84	75.9	372	0.654	0.190	0.025	0.066	0.039	0.018	0.009
12.00	6.73	75.6	331	0.669	0.188	0.022	0.066	0.033	0.016	0.005
13.00	6.84	66.8	241	0.675	0.191	0.023	0.064	0.032	0.012	0.003
15.00	6.86	65.6	155	0.707	0.186	0.020	0.056	0.023	0.007	0.001
17.00	6.92	57.5	127	0.719	0.178	0.020	0.051	0.024	0.006	0.001
19.00	7.09	46.2	119	0.718	0.179	0.021	0.048	0.027	0.006	0.001
21.00	7.21	46.8	85	0.724	0.179	0.022	0.042	0.026	0.006	0.001

Table 97

Composition of Rumen Contents of Sheep No. 449 on Treated Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.05	53.2	140	0.734	0.164	0.020	0.044	0.033	0.005	0.001
10.00	6.83	97.1	384	0.684	0.141	0.020	0.100	0.032	0.013	0.010
11.00	6.71	101.9	379	0.686	0.135	0.022	0.096	0.033	0.016	0.012
12.00	6.61	92.8	342	0.692	0.145	0.023	0.087	0.029	0.015	0.010
13.00	6.60	81.7	262	0.699	0.149	0.022	0.083	0.025	0.013	0.009
15.00	6.51	81.1	150	0.722	0.159	0.017	0.070	0.019	0.009	0.004
17.00	6.64	75.4	113	0.741	0.163	0.015	0.058	0.015	0.007	0.002
19.00	6.90	68.8	108	0.741	0.166	0.016	0.050	0.016	0.007	0.002
21.00	7.05	60.4	117	0.737	0.170	0.017	0.051	0.019	0.006	0.001

Table 98

Composition of Rumen Contents of Sheep No. 449 on Dried Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.94	740	239	0.704	0.172	0.023	0.066	0.028	0.008	0.002
10.00	6.70	98.5	461	0.732	0.156	0.017	0.064	0.021	0.008	0.002
11.00	6.60	111.9	530	0.737	0.152	0.015	0.066	0.018	0.010	0.001
12.00	6.58	105.3	394	0.726	0.159	0.015	0.071	0.017	0.010	0.001
13.00	6.64	90.1	334	0.719	0.165	0.016	0.072	0.017	0.010	0.002
15.00	6.60	89.9	265	0.703	0.175	0.017	0.073	0.021	0.010	0.002
17.00	6.69	79.7	222	0.714	0.172	0.015	0.067	0.020	0.010	0.002
19.00	6.54	77.2	216	0.719	0.170	0.016	0.064	0.020	0.009	0.002
21.00	6.75	75.7	237	0.720	0.166	0.018	0.061	0.024	0.009	0.001

Table 99

Composition of Blood from Individual Sheep

Metabolite	Treatment	Sheep No.								
		409	414	434	435	437	443	447	448	449
Blood pH	A	7.38	7.36	7.28	7.29	7.35	7.33	7.28	7.36	7.35
	B	7.38	7.28	7.35	7.35	7.36	7.41	7.34	7.28	7.38
	C	7.33	7.33	7.32	7.32	7.40	7.40	7.43	7.33	7.35
Plasma glucose mg/l	A	562	602	587	556	532	496	614	517	496
	B	532	547	617	614	547	466	617	575	556
	C	578	623	623	638	569	611	608	608	562
Plasma urea-nitrogen mg/l	A	229	272	222	190	217	212	200	262	178
	B	226	289	241	255	205	284	219	267	260
	C	260	277	212	190	262	226	270	250	341

A = Control silage
B = Treated silage
C = Dried grass

Table 100

Composition of Fresh Grass (Experiment 3A)

Date	Dry matter g/kg	g/kg dry matter			
		Crude protein	MAD-fibre	Ash	WSC(1)
15 May	207	104	229	80	290
16 May	226	105	232	79	266
17 May	208	97	262	81	270
18 May	193	97	241	70	282
19 May	207	89	255	65	240
Sampling day 1	219	91	260	66	238
21 May	202	92	268	74	253
Sampling day 2	203	90	259	63	290

(1) WSC = water soluble carbohydrate.

Table 101

Composition of Fresh Grass (Experiment 3B)

Date	Dry matter g/kg	g/kg dry matter			
		Crude protein	MAD-fibre	Ash	WSC
14 August	117	235	320	118	66
15 August	135	195	333	105	88
16 August	137	194	319	106	87
17 August	143	203	314	105	82
18 August	129	236	314	109	92
Sampling day 1	132	239	310	108	80
20 August	135	222	305	106	78
Sampling day 2	129	232	288	104	75

Table 102

Composition of Rumen Contents of Sheep No. 414 on Fresh Grass Diet (Experiment 3A)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions		
				C_2	C_3	nC_4
09.00	7.01	57.3	95	0.685	0.192	0.104
10.00	6.07	100.5	144	0.595	0.216	0.154
11.00	6.09	105.0	128	0.563	0.227	0.174
12.00	6.29	96.9	66	0.600	0.203	0.164
13.00	6.15	100.6	58	0.637	0.186	0.147
15.00	6.54	84.7	54	0.656	0.177	0.137
17.00	6.77	69.3	54	0.677	0.172	0.120
19.00	6.93	62.2	49	0.686	0.175	0.106
21.00	7.08	55.2	58	0.697	0.172	0.096

Table 103

Composition of Rumen Contents of Sheep No. 434 on Fresh Grass Diet (Experiment 3A)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions		
				C_2	C_3	nC_4
09.00	6.87	63.3	107	0.620	0.194	0.144
10.00	5.57	83.9	157	0.595	0.197	0.180
11.00	5.53	85.9	256	0.495	0.249	0.225
12.00	6.00	96.8	282	0.475	0.259	0.223
13.00	6.13	112.0	223	0.503	0.226	0.219
15.00	6.29	89.4	99	0.566	0.204	0.185
17.00	6.55	80.6	74	0.609	0.194	0.160
19.00	6.40	82.6	62	0.628	0.198	0.143
21.00	6.64	75.0	87	0.625	0.200	0.136

Table 104

Composition of Rumen Contents of Sheep No. 437 on Fresh Grass Diet (Experiment 3A)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions		
				C_2	C_3	nC_4
09.00	6.70	85.5	132	0.659	0.187	0.115
10.00	5.91	80.6	140	0.574	0.208	0.182
11.00	5.66	111.2	178	0.555	0.226	0.182
12.00	5.42	138.4	252	0.557	0.213	0.190
13.00	5.68	121.1	177	0.587	0.200	0.171
15.00	5.69	119.9	83	0.606	0.197	0.157
17.00	5.96	120.8	79	0.612	0.187	0.154
19.00	5.78	118.3	120	0.631	0.187	0.143
21.00	6.34	98.5	103	0.650	0.184	0.125

Table 105

Composition of Rumen Contents of Sheep No. 680 on Fresh Grass Diet (Experiment 3A)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions		
				C_2	C_3	nC_4
09.00	6.75	78.2	103	0.682	0.153	0.127
10.00	5.84	85.8	161	0.587	0.171	0.211
11.00	5.83	111.7	251	0.526	0.190	0.263
12.00	5.90	120.4	228	0.524	0.193	0.245
13.00	5.99	104.2	190	0.564	0.179	0.216
15.00	6.13	99.2	120	0.616	0.170	0.178
17.00	6.18	99.1	70	0.633	0.166	0.161
19.00	6.32	97.6	74	0.673	0.155	0.137
21.00	6.40	91.6	70	0.683	0.157	0.125

Table 106

Composition of Rumen Contents of Sheep No. 414 on Fresh Grass Diet (Experiment 3B)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions		
				C_2	C_3	nC_4
09.00	7.15	46.2	181	0.699	0.173	0.062
10.00	6.87	47.0	293	0.712	0.167	0.058
11.00	6.78	73.1	388	0.738	0.153	0.048
12.00	6.83	62.3	389	0.725	0.160	0.058
13.00	6.88	57.0	311	0.721	0.163	0.060
15.00	6.73	66.0	225	0.714	0.171	0.062
17.00	6.79	62.0	182	0.713	0.172	0.068
19.00	6.72	56.1	181	0.706	0.172	0.069
21.00	7.13	48.3	175	0.705	0.176	0.067

Table 107

Composition of Rumen Contents of Sheep No. 434 on Fresh Grass Diet (Experiment 3B)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions		
				C_2	C_3	nC_4
09.00	6.96	30.8	211	0.671	0.178	0.078
10.00	6.76	50.0	354	0.727	0.159	0.049
11.00	6.70	50.8	341	0.729	0.164	0.058
12.00	6.67	55.0	345	0.710	0.170	0.060
13.00	6.60	54.6	319	0.705	0.176	0.064
15.00	6.74	47.6	255	0.708	0.170	0.066
17.00	6.85	40.4	212	0.697	0.171	0.068
19.00	7.01	32.1	186	0.679	0.178	0.067
21.00	7.06	29.6	184	0.687	0.172	0.065

Table 108

Composition of Rumen Contents of Sheep No. 437 on Fresh Grass Diet (Experiment 3B)

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions		
				C_2	C_3	nC_4
09.00	6.86	57.7	152	0.690	0.201	0.059
10.00	6.76	60.9	336	0.706	0.182	0.047
11.00	6.54	69.0	367	0.735	0.183	0.051
12.00	6.48	82.5	423	0.708	0.189	0.059
13.00	6.46	78.2	302	0.702	0.189	0.060
15.00	6.42	72.7	259	0.700	0.196	0.063
17.00	6.60	68.2	225	0.692	0.200	0.061
19.00	6.76	67.0	233	0.685	0.203	0.062
21.00	6.78	61.3	221	0.685	0.203	0.062

Table 109

Nutritive Value of Silages and Grass for Individual Sheep

	Treatment	Sheep No.							
		409	414	433	435	436	437	680	V71
Daily intake g/kg $W^{0.75}$	A	25.9	22.3	33.8	7.7	19.2	31.5	33.8	32.2
	B	14.7	34.3	38.2	22.2	20.8	38.6	37.0	31.0
	C	42.4	27.4	36.3	14.4	28.7	41.4	39.7	30.8
	D	28.2	16.4	30.2	14.4	13.6	27.7	25.3	24.8
Daily intake g/day	A	566	455	723	156	306	650	587	851
	B	370	719	786	342	295	771	758	784
	C	960	599	763	223	472	952	756	813
	D	629	293	568	203	191	518	467	603
Dry matter digestibility	A	0.750	0.799	0.723	0.748	0.797	0.758	0.720	0.744
	B	0.825	0.764	0.756	0.781	0.819	0.761	0.762	0.781
	C	0.741	0.777	0.753	0.782	0.761	0.737	0.779	0.758
	D	0.796	0.806	0.763	0.852	0.847	0.798	0.814	0.794
Organic matter digestibility	A	0.765	0.816	0.739	0.795	0.813	0.772	0.736	0.758
	B	0.835	0.778	0.764	0.801	0.832	0.773	0.774	0.793
	C	0.750	0.791	0.762	0.799	0.776	0.751	0.790	0.769
	D	0.809	0.821	0.763	0.868	0.863	0.812	0.826	0.810
Nitrogen digestibility	A	0.729	0.759	0.725	0.717	0.757	0.741	0.691	0.787
	B	0.795	0.742	0.729	0.719	0.785	0.736	0.745	0.746
	C	0.761	0.762	0.783	0.755	0.757	0.743	0.796	0.758
	D	0.796	0.806	0.756	0.827	0.804	0.798	0.823	0.730

Table 109 (Cont'd)

Nutritive Value of Silages and Grass for Individual Sheep

	Treatment	Sheep No.							
		409	414	433	435	436	437	680	V71
Metabolisable energy MJ/kg	A	11.4	12.1	10.7	11.1	11.5	11.7	11.0	14.6
	B	12.2	11.7	11.4	10.6	12.3	11.4	11.8	11.7
	C	11.3	12.0	11.6	11.3	11.6	11.5	12.3	11.5
	D	13.8	13.9	12.8	14.4	13.3	13.6	13.9	12.2

A = Wilted silage
B = Grass
C = Acid treated wilted silage
D = Fresh silage

Table 110

Composition of Rumen Contents of Sheep No. 409 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.89	65.5	107	0.714	0.180	0.012	0.075	0.011	0.007	0.001
10.00	6.50	109.2	292	0.547	0.252	0.010	0.155	0.016	0.017	0.003
11.00	6.39	108.8	333	0.563	0.235	0.014	0.140	0.023	0.021	0.004
12.00	6.51	105.2	230	0.600	0.198	0.012	0.146	0.020	0.020	0.004
13.00	6.50	96.0	123	0.637	0.184	0.012	0.132	0.015	0.017	0.003
15.00	6.43	92.8	91	0.685	0.175	0.009	0.106	0.011	0.012	0.003
17.00	6.65	88.3	74	0.713	0.171	0.010	0.086	0.009	0.009	0.002
19.00	6.74	80.9	86	0.736	0.170	0.010	0.067	0.009	0.007	0.001
21.00	6.86	70.9	83	0.730	0.174	0.012	0.065	0.012	0.007	0.001

Table 111

Composition of Rumen Contents of Sheep No. 409 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.12	30.5	83	0.774	0.142	0.024	0.029	0.026	0.006	0.000
10.00	6.98	31.1	136	0.739	0.159	0.024	0.045	0.025	0.008	0.001
11.00	6.88	35.7	140	0.756	0.162	0.019	0.052	0.022	0.008	0.001
12.00	6.91	32.2	136	0.750	0.163	0.019	0.037	0.022	0.008	0.001
13.00	6.82	31.9	111	0.745	0.156	0.021	0.045	0.022	0.007	0.004
15.00	6.87	32.4	124	0.759	0.155	0.019	0.041	0.019	0.006	0.001
17.00	6.89	28.6	99	0.771	0.151	0.018	0.036	0.019	0.005	0.001
19.00	6.95	28.1	132	0.776	0.149	0.019	0.031	0.019	0.005	0.001
21.00	6.98	27.8	96	0.780	0.149	0.019	0.027	0.020	0.004	0.000

Table 112

Composition of Rumen Contents of Sheep No. 409 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.61	90.5	103	0.735	0.154	0.008	0.080	0.011	0.009	0.003
10.00	6.19	128.8	375	0.629	0.180	0.012	0.128	0.022	0.020	0.009
11.00	6.01	145.9	351	0.638	0.173	0.012	0.127	0.017	0.023	0.010
12.00	6.12	105.9	274	0.646	0.169	0.013	0.123	0.015	0.025	0.010
13.00	6.11	117.1	174	0.680	0.160	0.010	0.112	0.011	0.020	0.007
15.00	6.20	96.8	97	0.707	0.159	0.008	0.099	0.008	0.014	0.005
17.00	6.45	88.3	81	0.726	0.156	0.008	0.089	0.007	0.010	0.004
19.00	6.53	91.6	73	0.732	0.158	0.008	0.081	0.008	0.009	0.003
21.00	6.55	84.6	85	0.732	0.162	0.010	0.077	0.009	0.008	0.003

Table 113

Composition of Rumen Contents of Sheep No. 409 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.03	55.5	99	0.745	0.163	0.014	0.055	0.014	0.008	0.002
10.00	6.36	92.7	267	0.620	0.239	0.016	0.076	0.021	0.020	0.007
11.00	6.50	82.9	271	0.621	0.236	0.013	0.078	0.021	0.023	0.009
12.00	6.60	75.5	218	0.646	0.219	0.013	0.078	0.018	0.018	0.007
13.00	6.55	71.5	152	0.680	0.198	0.012	0.074	0.015	0.016	0.005
15.00	6.53	73.2	99	0.721	0.179	0.011	0.065	0.011	0.012	0.003
17.00	6.60	69.2	78	0.743	0.167	0.010	0.058	0.010	0.009	0.002
19.00	6.75	62.5	83	0.746	0.167	0.011	0.054	0.011	0.008	0.003
21.00	6.97	54.0	70	0.752	0.163	0.013	0.050	0.013	0.007	0.002

Table 114

Composition of Rumen Contents of Sheep No. 414 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.98	49.8	99	0.732	0.170	0.015	0.054	0.019	0.008	0.001
10.00	6.39	90.9	255	0.608	0.228	0.012	0.102	0.023	0.021	0.006
11.00	6.35	102.8	218	0.628	0.209	0.013	0.099	0.022	0.021	0.007
12.00	6.28	111.3	156	0.668	0.192	0.010	0.091	0.017	0.018	0.004
13.00	6.53	76.4	115	0.674	0.193	0.013	0.086	0.016	0.014	0.003
15.00	6.65	78.3	74	0.709	0.180	0.010	0.076	0.011	0.011	0.003
17.00	6.72	70.9	78	0.728	0.174	0.012	0.064	0.013	0.008	0.002
19.00	6.84	53.0	82	0.736	0.172	0.014	0.055	0.015	0.007	0.002
21.00	6.93	51.8	115	0.738	0.170	0.014	0.052	0.017	0.007	0.001

Table 115

Composition of Rumen Contents of Sheep No. 414 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.84	57.8	156	0.714	0.163	0.015	0.078	0.017	0.009	0.004
10.00	6.50	88.2	235	0.654	0.220	0.010	0.085	0.013	0.012	0.006
11.00	6.50	96.2	177	0.658	0.187	0.010	0.077	0.034	0.030	0.004
12.00	6.41	92.9	136	0.695	0.184	0.010	0.082	0.013	0.012	0.005
13.00	6.44	85.6	95	0.712	0.173	0.009	0.080	0.011	0.011	0.004
15.00	6.58	81.3	86	0.731	0.165	0.009	0.073	0.011	0.009	0.003
17.00	6.68	75.0	144	0.731	0.164	0.011	0.071	0.013	0.008	0.003
19.00	6.79	73.6	152	0.736	0.159	0.013	0.066	0.016	0.008	0.002
21.00	6.90	64.2	173	0.731	0.159	0.015	0.066	0.019	0.008	0.002

Table 116

Composition of Rumen Contents of Sheep No. 414 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.09	54.7	136	0.732	0.155	0.015	0.070	0.019	0.008	0.002
10.00	6.83	88.5	296	0.621	0.218	0.012	0.106	0.020	0.020	0.004
11.00	6.64	91.0	267	0.644	0.187	0.014	0.104	0.023	0.023	0.005
12.00	6.74	80.2	165	0.688	0.178	0.014	0.073	0.018	0.018	0.011
13.00	6.74	76.6	107	0.689	0.169	0.014	0.094	0.016	0.014	0.003
15.00	6.81	68.7	82	0.722	0.161	0.012	0.079	0.013	0.010	0.002
17.00	6.98	52.1	87	0.746	0.150	0.013	0.070	0.013	0.008	0.002
19.00	7.04	46.7	82	0.748	0.150	0.014	0.066	0.014	0.007	0.001
21.00	6.98	43.2	96	0.740	0.153	0.016	0.065	0.017	0.007	0.001

Table 117

Composition of Rumen Contents of Sheep No. 414 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.10	48.1	156	0.733	0.184	0.015	0.043	0.018	0.006	0.001
10.00	6.69	71.4	177	0.666	0.235	0.013	0.049	0.021	0.014	0.003
11.00	6.63	76.3	166	0.671	0.227	0.012	0.056	0.017	0.014	0.004
12.00	6.70	71.0	137	0.687	0.219	0.011	0.056	0.015	0.011	0.002
13.00	6.69	64.0	89	0.705	0.212	0.012	0.050	0.012	0.009	0.002
15.00	6.78	61.9	81	0.730	0.196	0.011	0.045	0.010	0.007	0.002
17.00	6.83	51.4	81	0.738	0.190	0.010	0.045	0.010	0.006	0.001
19.00	6.92	50.0	85	0.736	0.188	0.012	0.043	0.012	0.008	0.002
21.00	7.03	49.3	89	0.738	0.186	0.014	0.040	0.015	0.006	0.001

Table 118

Composition of Rumen Contents of Sheep No. 433 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.86	62.5	70	0.719	0.181	0.011	0.070	0.013	0.006	0.001
10.00	6.32	121.5	267	0.607	0.279	0.009	0.075	0.016	0.013	0.001
11.00	6.36	112.3	371	0.595	0.293	0.009	0.073	0.017	0.013	0.000
12.00	6.35	121.9	387	0.600	0.270	0.012	0.080	0.019	0.018	0.002
13.00	6.51	106.5	272	0.630	0.240	0.012	0.081	0.017	0.019	0.002
15.00	6.48	94.9	169	0.656	0.225	0.010	0.080	0.014	0.014	0.001
17.00	6.39	75.1	111	0.678	0.209	0.013	0.078	0.012	0.010	0.000
19.00	6.64	67.0	78	0.710	0.189	0.011	0.070	0.010	0.011	0.001
21.00	6.73	62.3	74	0.725	0.178	0.011	0.067	0.012	0.006	0.001

Table 119

Composition of Rumen Contents of Sheep No. 433 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.67	67.3	95	0.675	0.217	0.012	0.074	0.010	0.009	0.002
10.00	6.31	72.1	181	0.634	0.279	0.007	0.061	0.008	0.009	0.002
11.00	6.07	95.5	194	0.607	0.288	0.008	0.072	0.009	0.012	0.004
12.00	6.36	95.2	153	0.614	0.280	0.008	0.072	0.008	0.014	0.004
13.00	5.93	108.9	166	0.618	0.272	0.007	0.079	0.007	0.013	0.005
15.00	6.48	86.9	123	0.658	0.234	0.008	0.076	0.008	0.013	0.004
17.00	6.43	79.2	109	0.655	0.230	0.009	0.079	0.010	0.013	0.004
19.00	6.49	74.8	97	0.664	0.225	0.009	0.077	0.010	0.013	0.003
21.00	6.45	62.1	101	0.710	0.198	0.012	0.057	0.013	0.009	0.002

Table 120

Composition of Rumen Contents of Sheep No. 433 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.01	53.0	103	0.681	0.208	0.016	0.069	0.018	0.007	0.001
10.00	6.20	110.6	329	0.576	0.326	0.009	0.057	0.016	0.014	0.002
11.00	6.38	88.8	337	0.574	0.319	0.010	0.060	0.019	0.017	0.001
12.00	6.43	95.4	345	0.579	0.296	0.015	0.066	0.020	0.021	0.002
13.00	6.05	126.7	280	0.597	0.277	0.013	0.071	0.017	0.021	0.003
15.00	6.61	71.9	148	0.635	0.249	0.014	0.076	0.013	0.012	0.002
17.00	6.59	81.2	99	0.668	0.224	0.013	0.072	0.013	0.009	0.001
19.00	6.68	76.1	78	0.680	0.213	0.013	0.070	0.014	0.008	0.003
21.00	6.88	53.9	95	0.689	0.208	0.014	0.065	0.016	0.007	0.002

Table 121

Composition of Rumen Contents of Sheep No. 433 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.85	38.2	95	0.716	0.182	0.015	0.067	0.012	0.006	0.001
10.00	6.38	76.7	247	0.627	0.255	0.009	0.077	0.018	0.012	0.002
11.00	6.42	90.9	283	0.628	0.242	0.011	0.081	0.021	0.015	0.003
12.00	6.51	82.2	247	0.636	0.227	0.012	0.084	0.023	0.015	0.003
13.00	6.55	73.8	205	0.664	0.212	0.012	0.080	0.019	0.013	0.002
15.00	6.67	59.9	156	0.695	0.200	0.011	0.070	0.014	0.008	0.001
17.00	6.75	49.6	106	0.714	0.191	0.011	0.065	0.012	0.007	0.001
19.00	6.78	42.6	87	0.723	0.184	0.010	0.065	0.011	0.006	0.001
21.00	6.78	35.8	83	0.733	0.176	0.013	0.061	0.011	0.005	0.001

Table 122

Composition of Rumen Contents of Sheep No. 435 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.26	27.3	66	0.706	0.183	0.023	0.058	0.021	0.008	0.002
10.00	7.01	48.8	132	0.641	0.214	0.020	0.081	0.025	0.018	0.002
11.00	7.02	48.8	123	0.636	0.209	0.021	0.085	0.028	0.020	0.002
12.00	7.08	43.2	91	0.648	0.203	0.020	0.082	0.027	0.018	0.002
13.00	7.10	38.8	74	0.666	0.195	0.019	0.079	0.024	0.016	0.001
15.00	7.10	37.6	82	0.691	0.189	0.019	0.067	0.022	0.012	0.001
17.00	7.19	32.3	78	0.709	0.178	0.020	0.059	0.022	0.012	0.002
19.00	7.25	28.0	62	0.714	0.180	0.022	0.052	0.023	0.009	0.000
21.00	7.16	28.1	74	0.720	0.169	0.024	0.048	0.025	0.015	0.000

Table 123

Composition of Rumen Contents of Sheep No. 435 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.17	57.2	95	0.753	0.161	0.015	0.044	0.018	0.008	0.002
10.00	6.79	70.4	119	0.690	0.204	0.011	0.061	0.017	0.013	0.004
11.00	6.67	70.6	115	0.692	0.198	0.012	0.067	0.014	0.013	0.004
12.00	6.80	73.3	95	0.699	0.199	0.011	0.061	0.013	0.014	0.004
13.00	6.75	75.9	66	0.717	0.189	0.011	0.057	0.011	0.013	0.003
15.00	6.86	62.3	74	0.736	0.178	0.010	0.054	0.010	0.010	0.002
17.00	6.94	61.3	74	0.751	0.167	0.011	0.047	0.013	0.008	0.002
19.00	7.00	59.5	95	0.749	0.167	0.015	0.044	0.016	0.008	0.001
21.00	7.03	54.0	96	0.746	0.163	0.018	0.046	0.019	0.008	0.001

Table 124

Composition of Rumen Contents of Sheep No. 435 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.69	46.7	112	0.763	0.156	0.015	0.032	0.025	0.008	0.002
10.00	6.26	92.1	311	0.626	0.255	0.013	0.059	0.030	0.015	0.002
11.00	6.12	94.1	371	0.609	0.269	0.012	0.066	0.025	0.018	0.002
12.00	6.54	74.6	274	0.642	0.230	0.014	0.061	0.029	0.022	0.002
13.00	6.66	61.5	170	0.679	0.211	0.013	0.054	0.026	0.015	0.001
15.00	6.66	57.8	125	0.721	0.191	0.012	0.044	0.020	0.011	0.001
17.00	6.56	56.0	81	0.742	0.174	0.013	0.042	0.020	0.008	0.001
19.00	6.63	49.9	77	0.759	0.162	0.012	0.036	0.018	0.007	0.006
21.00	6.65	53.1	89	0.710	0.196	0.011	0.056	0.015	0.010	0.002

Table 125

Composition of Rumen Contents of Sheep No. 435 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.22	26.6	111	0.717	0.177	0.028	0.033	0.036	0.008	0.001
10.00	6.86	41.1	165	0.641	0.231	0.023	0.051	0.038	0.015	0.002
11.00	6.76	49.1	161	0.625	0.243	0.020	0.057	0.035	0.017	0.004
12.00	6.73	50.2	144	0.650	0.223	0.018	0.059	0.032	0.016	0.002
13.00	6.80	45.7	111	0.662	0.218	0.019	0.055	0.030	0.014	0.002
15.00	6.99	36.1	74	0.701	0.200	0.021	0.043	0.025	0.010	0.001
17.00	6.97	36.8	78	0.713	0.181	0.024	0.042	0.031	0.009	0.000
19.00	7.05	28.2	78	0.726	0.173	0.025	0.036	0.030	0.008	0.001
21.00	7.16	29.0	74	0.731	0.170	0.026	0.030	0.030	0.013	0.000

Table 126

Composition of Rumen Contents of Sheep No. 436 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.97	32.4	115	0.711	0.190	0.023	0.042	0.023	0.009	0.003
10.00	6.68	67.6	226	0.613	0.261	0.014	0.065	0.026	0.019	0.002
11.00	6.58	72.9	214	0.608	0.263	0.016	0.067	0.024	0.020	0.004
12.00	6.42	78.1	137	0.637	0.243	0.014	0.066	0.020	0.018	0.004
13.00	6.55	62.1	89	0.668	0.224	0.013	0.062	0.017	0.014	0.003
15.00	6.72	54.6	65	0.699	0.205	0.013	0.057	0.015	0.010	0.002
17.00	6.79	46.8	73	0.716	0.198	0.012	0.049	0,014	0.009	0.001
19.00	6.87	40.6	77	0.727	0.188	0.015	0.044	0.017	0.008	0.001
21.00	6.93	34.5	77	0.721	0.189	0.018	0.044	0.019	0.007	0.001

Table 127

Composition of Rumen Contents of Sheep No. 436 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.10	24.7	107	0.693	0.167	0.028	0.065	0.032	0.011	0.002
10.00	6.90	32.0	115	0.667	0.201	0.023	0.062	0.026	0.017	0.005
11.00	6.95	33.1	120	0.676	0.198	0.021	0.062	0.022	0.016	0.005
12.00	6.81	35.7	82	0.695	0.187	0.018	0.065	0.019	0.012	0.004
13.00	6.95	29.3	66	0.696	0.185	0.022	0.062	0.020	0.012	0.002
15.00	7.07	30.0	74	0.715	0.170	0.023	0.061	0.018	0.010	0.003
17.00	7.08	28.6	87	0.717	0.163	0.025	0.057	0.024	0.012	0.002
19.00	7.13	20.0	83	0.722	0.156	0.026	0.056	0.028	0.012	0.002
21.00	7.14	22.9	94	0.718	0.149	0.031	0.058	0.032	0.010	0.003

Table 128

Composition of Rumen Contents of Sheep No. 436 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.95	56.2	86	0.703	0.208	0.013	0.055	0.013	0.007	0.001
10.00	6.73	98.6	189	0.612	0.277	0.012	0.065	0.016	0.016	0.001
11.00	6.33	106.3	259	0.586	0.300	0.010	0.067	0.018	0.018	0.001
12.00	6.22	108.5	197	0.610	0.282	0.011	0.071	0.016	0.008	0.002
13.00	6.16	100.1	124	0.634	0.256	0.010	0.071	0.012	0.016	0.002
15.00	6.66	82.7	62	0.670	0.232	0.010	0.066	0.010	0.010	0.001
17.00	6.75	65.7	62	0.695	0.211	0.010	0.064	0.011	0.008	0.001
19.00	6.92	61.2	82	0.711	0.202	0.012	0.056	0.012	0.007	0.001
21.00	7.01	54.2	83	0.710	0.199	0.014	0.053	0.016	0.007	0.001

Table 129

Composition of Rumen Contents of Sheep No. 436 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	7.17	27.5	82	0.687	0.192	0.022	0.054	0.035	0.008	0.002
10.00	7.15	40.7	111	0.649	0.224	0.021	0.057	0.030	0.015	0.004
11.00	7.10	39.0	128	0.653	0.233	0.021	0.042	0.030	0.017	0.004
12.00	7.12	41.8	107	0.658	0.226	0.022	0.060	0.018	0.014	0.003
13.00	7.10	42.0	95	0.666	0.212	0.023	0.057	0.029	0.012	0.003
15.00	7.00	36.1	74	0.678	0.206	0.021	0.060	0.024	0.010	0.000
17.00	7.03	30.8	82	0.688	0.198	0.022	0.057	0.025	0.009	0.001
19.00	7.22	26.0	78	0.693	0.191	0.026	0.056	0.028	0.006	0.000
21.00	7.25	24.9	74	0.683	0.199	0.025	0.055	0.029	0.009	0.000

Table 130

Composition of Rumen Contents of Sheep No. 437 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.82	59.4	103	0.721	0.169	0.014	0.068	0.015	0.011	0.003
10.00	6.46	84.7	282	0.569	0.216	0.012	0.158	0.019	0.019	0.008
11.00	6.52	84.8	274	0.561	0.206	0.015	0.163	0.022	0.024	0.009
12.00	6.56	120.2	186	0.587	0.197	0.017	0.147	0.016	0.025	0.010
13.00	6.49	84.1	157	0.631	0.184	0.014	0.125	0.016	0.022	0.009
15.00	6.28	91.2	129	0.660	0.180	0.011	0.109	0.013	0.020	0.008
17.00	6.73	55.0	105	0.698	0.176	0.013	0.083	0.013	0.014	0.004
19.00	6.74	57.1	97	0.704	0.177	0.012	0.078	0.013	0.012	0.003
21.00	6.67	60.9	113	0.709	0.179	0.013	0.070	0.015	0.012	0.004

Table 131

Composition of Rumen Contents of Sheep No. 437 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.23	105.1	156	0.695	0.170	0.012	0.094	0.016	0.010	0.003
10.00	6.25	95.2	218	0.643	0.195	0.012	0.117	0.017	0.012	0.005
11.00	6.28	89.0	189	0.646	0.194	0.012	0.115	0.016	0.012	0.005
12.00	6.29	87.1	152	0.670	0.182	0.010	0.106	0.015	0.012	0.005
13.00	6.33	90.0	144	0.676	0.183	0.011	0.100	0.015	0.011	0.004
15.00	5.93	117.7	123	0.677	0.181	0.011	0.103	0.012	0.011	0.004
17.00	6.08	113.3	110	0.683	0.177	0.011	0.099	0.015	0.011	0.005
19.00	6.12	112.1	168	0.683	0.175	0.011	0.097	0.017	0.013	0.004
21.00	6.52	88.7	160	0.694	0.172	0.014	0.091	0.018	0.010	0.003

Table 132

Composition of Rumen Contents of Sheep No. 437 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.61	81.5	82	0.693	0.207	0.010	0.072	0.010	0.008	0.001
10.00	6.04	129.1	271	0.600	0.284	0.008	0.081	0.014	0.012	0.001
11.00	6.18	125.9	325	0.581	0.273	0.009	0.101	0.019	0.016	0.002
12.00	5.95	137.1	317	0.601	0.252	0.010	0.099	0.018	0.019	0.002
13.00	6.03	114.9	280	0.619	0.238	0.011	0.097	0.015	0.019	0.002
15.00	5.97	117.6	120	0.657	0.226	0.008	0.086	0.011	0.013	0.001
17.00	6.35	93.8	99	0.678	0.213	0.008	0.079	0.012	0.010	0.001
19.00	6.53	89.1	91	0.694	0.204	0.008	0.073	0.011	0.009	0.001
21.00	6.64	77.8	83	0.694	0.204	0.010	0.071	0.012	0.008	0.001

Table 133

Composition of Rumen Contents of Sheep No. 437 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.90	47.6	115	0.735	0.176	0.012	0.051	0.018	0.006	0.001
10.00	6.74	70.0	214	0.647	0.219	0.012	0.083	0.024	0.013	0.002
11.00	6.59	74.9	231	0.646	0.229	0.012	0.068	0.029	0.014	0.003
12.00	6.78	64.3	181	0.656	0.211	0.012	0.083	0.026	0.011	0.002
13.00	6.69	58.2	120	0.680	0.199	0.012	0.076	0.023	0.009	0.002
15.00	6.66	48.4	91	0.708	0.191	0.010	0.063	0.021	0.007	0.001
17.00	6.50	56.9	90	0.727	0.182	0.011	0.057	0.017	0.007	0.001
19.00	6.79	47.0	86	0.732	0.180	0.012	0.051	0.019	0.006	0.001
21.00	6.83	47.1	118	0.739	0.177	0.014	0.045	0.019	0.006	0.001

Table 134

Composition of Rumen Contents of Sheep No. 680 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.94	59.5	99	0.735	0.168	0.014	0.057	0.016	0.008	0.002
10.00	6.35	94.3	292	0.602	0.205	0.014	0.129	0.023	0.020	0.007
11.00	6.37	103.1	329	0.592	0.204	0.014	0.141	0.018	0.023	0.008
12.00	6.47	88.1	226	0.635	0.194	0.012	0.119	0.017	0.018	0.005
13.00	6.49	83.2	152	0.661	0.187	0.012	0.106	0.015	0.015	0.004
15.00	6.64	73.7	99	0.700	0.175	0.013	0.087	0.013	0.010	0.003
17.00	6.50	68.8	82	0.724	0.168	0.011	0.072	0.012	0.009	0.003
19.00	6.65	72.0	87	0.738	0.166	0.013	0.060	0.013	0.008	0.003
21.00	6.80	63.9	86	0.741	0.164	0.013	0.059	0.014	0.007	0.002

Table 135

Composition of Rumen Contents of Sheep No. 680 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.69	74.6	156	0.710	0.176	0.014	0.077	0.013	0.011	0.001
10.00	6.30	98.6	255	0.643	0.233	0.009	0.087	0.012	0.011	0.006
11.00	6.24	103.8	292	0.655	0.211	0.009	0.090	0.015	0.014	0.006
12.00	6.19	96.7	214	0.674	0.199	0.009	0.087	0.013	0.013	0.006
13.00	6.33	91.7	189	0.683	0.193	0.009	0.088	0.012	0.011	0.004
15.00	6.13	100.6	144	0.701	0.187	0.008	0.080	0.010	0.010	0.004
17.00	6.07	105.3	124	0.727	0.166	0.007	0.076	0.010	0.010	0.004
19.00	6.24	106.8	123	0.714	0.184	0.008	0.071	0.011	0.009	0.004
21.00	6.43	96.5	144	0.713	0.175	0.010	0.077	0.013	0.009	0.003

Table 136

Composition of Rumen Contents of Sheep No. 680 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.67	87.9	123	0.712	0.172	0.010	0.077	0.015	0.010	0.004
10.00	6.33	117.1	329	0.608	0.274	0.006	0.080	0.018	0.012	0.002
11.00	6.23	126.9	366	0.605	0.286	0.010	0.060	0.019	0.019	0.002
12.00	6.39	132.0	395	0.619	0.248	0.011	0.082	0.017	0.020	0.003
13.00	6.45	109.3	284	0.646	0.231	0.010	0.079	0.016	0.018	0.002
15.00	6.22	103.1	177	0.693	0.205	0.008	0.073	0.011	0.011	0.001
17.00	6.18	99.2	111	0.703	0.193	0.008	0.076	0.010	0.009	0.001
19.00	6.45	91.3	82	0;733	0.172	0.007	0.066	0.010	0.010	0.001
21.00	6.50	86.3	80	0.731	0.174	0.008	0.068	0.012	0.006	0.001

Table 137

Composition of Rumen Contents of Sheep No. 680 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.83	38.6	121	0.733	0.170	0.016	0.049	0.021	0.009	0.002
10.00	6.36	73.6	250	0.619	0.245	0.013	0.074	0.027	0.018	0.004
11.00	6.37	73.9	266	0.609	0.247	0.014	0.078	0.028	0.019	0.005
12.00	6.36	74.3	206	0.635	0.225	0.013	0.079	0.026	0.018	0.004
13.00	6.65	53.9	170	0.655	0.216	0.014	0.074	0.025	0.014	0.003
15.00	6.60	50.8	129	0.698	0.195	0.012	0.063	0.019	0.011	0.002
17.00	6.65	40.9	113	0.704	0.191	0.013	0.060	0.021	0.010	0.002
19.00	6.76	33.6	109	0.720	0.184	0.013	0.051	0.019	0.009	0.005
21.00	6.63	46.4	133	0.716	0.194	0.014	0.049	0.019	0.008	0.001

Table 138

Composition of Rumen Contents of Sheep No. V71 on Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.73	77.2	95	0.741	0.162	0.009	0.064	0.011	0.009	0.004
10.00	6.65	106.8	333	0.584	0.223	0.010	0.144	0.019	0.016	0.005
11.00	6.54	102.0	387	0.596	0.233	0.013	0.111	0.020	0.020	0.008
12.00	6.51	114.2	337	0.611	0.202	0.012	0.131	0.016	0.021	0.007
13.00	6.55	101.0	198	0.649	0.190	0.012	0.116	0.013	0.016	0.005
15.00	6.40	78.9	120	0.700	0.178	0.006	0.090	0.009	0.011	0.004
17.00	6.67	78.2	78	0.730	0.171	0.008	0.072	0.008	0.008	0.002
19.00	6.70	73.2	83	0.746	0.171	0.007	0.060	0.007	0.007	0.002
21.00	6.73	75.9	92	0.763	0.160	0.008	0.053	0.008	0.007	0.001

Table 139

Composition of Rumen Contents of Sheep No. V71 on Grass Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.54	77.7	186	0.712	0.169	0.014	0.071	0.023	0.010	0.003
10.00	6.31	85.4	279	0.633	0.230	0.010	0.091	0.018	0.012	0.005
11.00	6.38	80.5	226	0.651	0.207	0.013	0.092	0.019	0.013	0.004
12.00	6.42	91.7	259	0.678	0.190	0.012	0.085	0.018	0.012	0.004
13.00	6.39	82.8	153	0.682	0.188	0.011	0.087	0.018	0,012	0.002
15.00	6.34	86.4	145	0.694	0.179	0.011	0.085	0.017	0.011	0.003
17.00	6.30	86.5	141	0.696	0.177	0.012	0.084	0.017	0.011	0.004
19.00	6.36	87.2	133	0.712	0.171	0.012	0.076	0.018	0.010	0.003
21.00	6.34	91.1	137	0.716	0.165	0.012	0.076	0.019	0.009	0.003

Table 140

Composition of Rumen Contents of Sheep No. V71 on Acid Treated Wilted Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.58	74.4	124	0.726	0.167	0.011	0.071	0.013	0.010	0.003
10.00	6.38	99.1	275	0.629	0.174	0.013	0.138	0.022	0.018	0.007
11.00	6.45	102.8	383	0.612	0.171	0.013	0.151	0.021	0.023	0.009
12.00	6.46	99.5	316	0.648	0.166	0.013	0.130	0.017	0.020	0.008
13.00	6.40	90.0	226	0.666	0.165	0.011	0.121	0.012	0.017	0.007
15.00	6.43	100.3	156	0.707	0.164	0.009	0.093	0.011	0.012	0.004
17.00	6.51	84.3	119	0.722	0.165	0.009	0.079	0.010	0.010	0.005
19.00	6.41	84.0	95	0.734	0.168	0.009	0.069	0.010	0.008	0.002
21.00	6.48	89.8	140	0.737	0.170	0.009	0.064	0.011	0.008	0.003

Table 141

Composition of Rumen Contents of Sheep No. V71 on Fresh Silage Diet

Time of sampling (h)	pH	TVFA m mol/l	NH_3-N mg/l	Molar proportions						
				C_2	C_3	iC_4	nC_4	iC_5	nC_5	nC_6
09.00	6.81	61.0	132	0.720	0.163	0.012	0.085	0.013	0.006	0.001
10.00	6.55	86.3	284	0.655	0.229	0.011	0.065	0.023	0.014	0.003
11.00	6.51	94.0	300	0.643	0.233	0.013	0.068	0.025	0.015	0.004
12.00	6.72	74.7	251	0.673	0.220	0.011	0.061	0.021	0.012	0.002
13.00	6.62	70.4	210	0.686	0.209	0.011	0.062	0.019	0.010	0.002
15.00	6.72	70.1	111	0.731	0.187	0.009	0.051	0.014	0.007	0.001
17.00	6.78	63.5	136	0.752	0.176	0.009	0.040	0.013	0.009	0.001
19.00	6.75	63.3	128	0.760	0.168	0.011	0.043	0.013	0.006	0.000
21.00	6.85	54.6	131	0.767	0.163	0.012	0.039	0.014	0.005	0.000

Table 142

Composition of Blood from Individual Sheep

Metabolite	Treatment	Sheep No.							
		409	414	433	435	436	437	680	V71
Blood pH	A	7.34	7.31	7.36	7.28	7.25	7.31	7.24	7.16
	B	7.41	7.37	7.28	7.37	7.19	7.37	7.40	7.25
	C	7.33	7.33	7.36	7.26	7.33	7.39	7.31	7.28
	D	7.39	7.16	7.35	7.22	7.15	7.28	7.27	7.30
Plasma glucose mg/l	A	529	558	676	472	594	552	579	585
	B	579	629	594	518	588	-	584	558
	C	533	567	646	482	608	614	479	606
	D	494	564	687	521	567	558	467	562
Plasma urea-nitrogen mg/l	A	175	218	169	138	150	146	197	169
	B	148	155	143	176	174	143	121	164
	C	197	197	155	92	177	126	151	169
	D	101	185	121	150	139	134	143	182